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SREDOZEMSKI MORSKI PSI

SQUALI MEDITERRANEI

MEDITERRANEAN SHARKS

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PROVOKED NON-FATAL ATTACKS TO DIVERS BY SANDBAR SHARK, *CARCHARHINUS PLUMBEUS* (CARCHARHINIFORMES: CARCHARHINIDAE), OFF TAŞUCU COAST (NE MEDITERRANEAN SEA, TURKEY)

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ABSTRACT

On 26 August 2019, two commercial divers, who were diving for the routine check and cleaning of the separate aquaculture net cages, were attacked by several sandbar sharks, Carcharhinus plumbeus. *Seven to 8 specimens of sharks attacked the divers, while they were cleaning the entangled dead farmed fishes from outside of the cages. Although, the sea bottom depth, where the aquaculture cages are anchored, is from 47 to 68 m, the incidents happened in midwater around 20 m deep. The present incidents were apparently provoked by the presence of excess amount of wounded and/or dead farmed fish, which caused a certain feeding frenzy of a shark species, normally considered not to be particularly dangerous.*

Key words: Sandbar shark, Carcharhinidae, aquaculture, provoked attack, conservation, feeding frenzy

ATTACCHI NON FATALI PROVOCATI A SUBACQUEI DALLO SQUALO GRIGIO, *CARCHARHINUS PLUMBEUS* (CARCHARHINIFORMES: CARCHARHINIDAE), AL LARGO DELLA COSTA DI TAŞUCU (MEDITERRANEO NORD-ORIENTALE, TURCHIA)

SINTESI

Il 26 agosto 2019, due subacquei che si stavano immergendo per un controllo di routine e la pulizia delle gabbie di una rete di acquacoltura, sono stati attaccati da diversi squali grigi, Carcharhinus plumbeus. *Circa sette-otto esemplari hanno attaccato i subacquei, mentre stavano pulendo i pesci morti impigliati, dall'esterno delle gabbie d'allevamento. Sebbene il fondo del mare dove sono ancorate le gabbie di acquacoltura si trovi a 47-68 m di profondità, gli incidenti si sono verificati a 20 m di profondità circa. Questi incidenti sono stati apparentemente provocati dalla presenza di quantità eccessive di pesci d'allevamento feriti e/o morti, che hanno causato una certa frenesia alimentare ad una specie di squalo normalmente considerata non particolarmente pericolosa.*

Parole chiave: squalo grigio, Carcharhinidae, acquacoltura, attacco provocato, conservazione, frenesia alimentare

INTRODUCTION

Sharks as a group are considered to be highly successful predatory fishes, and are generally asynchronous opportunistic feeders on the most abundant prey item, which are primarily other fishes (Motta & Wilga, 2001). The presence of blood in the water, as from an injured organism in the sea, has long been regarded as a strong motivator for shark attack (Randall, 1986). Moreover, abundance of prey, blood, and irregular movements like those of a struggling fish, creating assorted and numerous stimuli in the water, can trigger a type of behavior known as a "feeding frenzy" (Springer & Gold, 1989).

Although, aquaculture offers great potential providing sustainable sources of food fish, interaction and compatibility of aquaculture with the environment, and vice versa, is one of the main debated issue (Massa *et al.*, 2017). Aggregation of wild fish nearby offshore aquaculture cages and the possibility of modifying the spatial and temporal extent of the aggregated fish is still a poorly understood phenomenon (Bacher *et al.*, 2012; Özgül & Angel, 2013). Aquaculture farms can attract predatory fish, like sharks, due to the presence of easy food opportunities in form of unconsumed feed and farmed fish (Papastimatiou *et al.*, 2010; Callier *et al.*, 2018).

In the present article, authors report on two incidents of provoked non-fatal shark attacks, occurred on 26 August 2019, nearby an aquaculture cage, off Taşucu coast (northeastern Mediterranean Sea, Turkey).

MATERIAL AND METHODS

Since every opportunity to examine a dead wild animal has some potential research value, the selection of an appropriate sample for the present study was an instance of typical opportunistic research, consisting in dead animal sampling (Jessup, 2003). On 26 August 2019, during a site survey in the vicinity of an aquaculture cage farm off Taşucu Dana Island coast (Fig. 1), the second author of the present article has interviewed with the employees of the fish farm and gathered information about the incident of a shark attack to two divers. The locality of the aquaculture cages is nearly 4 km off the coast and no human settlement is present in the vicinity of the farm area. Moreover, entering or trepassing the farm area is prohibited and subjected to permission. The mentioned information included some photographs of the shark on board of the support vessel and of close up photos of the lacerated diving gears (Figs. 2, 3 & 4), and a video footage with an approximately 3 minutes. The photograph seen in figure 2, which is



Fig. 1: Map depicting the approximate locality of the shark attack incident.
Sl. 1: Zemljevid obravnavanega območja z lokaliteto, kjer je prišlo do napadov morskih psov.



Fig. 2. One of the sandbar sharks,* C. plumbeus*, which attacked the diver under stimulated foraging conditions (Photo: İbrahim Yörüsün).
Sl. 2: Eden izmed sivih morskih psov,* C. plumbeus*, ki je vzdražen zaradi prisotnosti hrane napadel potapljača (Foto: İbrahim Yörüsün).

depicting the shark from a very clear side view, was used to confirm the identification of the species of the shark, following the descriptive criteria proposed by Serena (2005) and Ebert & Stehmann (2013).

RESULTS AND DISCUSSION

The shark species depicted in Figure 2 was identified as *Carcharhinus plumbeus* (Nardo, 1827). Although, not clearly seen on the photograph, an interdorsal ridge is present. First dorsal fin origin is over pectoral fin base, and first dorsal fin is extremely tall and semifalcate (Fig. 2). Total length of the examined sandbar shark was 2.3 m.

On 26 August 2019, two commercial divers, who were diving for the routine check and cleaning of the net cages, were attacked by several sharks. The net cages are used for the farming of European sea bass, *Dicentrarchus labrax* (Linnaeus, 1758). Numerous dead farmed fish were seen while they were floating in midwater or sank on the bottom of the cages. The following reconstruction of the two separate shark attacks is based on the individual testimonies of the divers 1 and 2, and the detailed examination of the video footage. The first diver dived into cage at a depth of 68 m; however, diver 1 stopped descending around 20 m deep, and started cleaning and routine maintenance work. While he was performing a routine maintenance dive, he suddenly felt a bump from below and a shark has bitten his diving boots and fins (Fig. 3). Diver 1 emphasized that he didn't see the shark was approaching him. The second diver has also descended to nearly 20 m deep in the vicinity of another cage, where depth of the bottom is 47 m. Diver 2 has also started his daily routine clearance and maintenance dive, outside the cage. Since the sharks were already in a frenzied status, a group of sandbar sharks approached him, started

Tab. 1: Shark attack incidents caused by the sandbar shark and logged in Global Shark Attack File (GSAF). All of the localities are along northwestern Atlantic coast.
Tab. 1: Primeri napadov sivega morskega psa iz globalne podatkovne baze o napadih morskih psov (GSAF). Vse lokalitete so vzdolž severozahodne atlantske obale.

GSAF log No	Date	Provoked	Unprovoked	Locality	Remarks (TL)
2601	22/6/1966	✓		New Jersey	2.1 m female
2654	Aug 1967		✓	Bahamas	1.5 to 1.8 m
3133	25/3/1981		✓	Florida	1.2 to 1.5 m
3501	09/9/1989		✓	North Carolina	Not available
4324	26/7/2002		✓	South Carolina	1.2 m
4606	21/8/2005		✓	South Carolina	Not available
5233	10/5/2011	✓		Florida	ca. 2.5 m
5608	09/7/2014		✓	Delaware	1.2 to 1.5 m

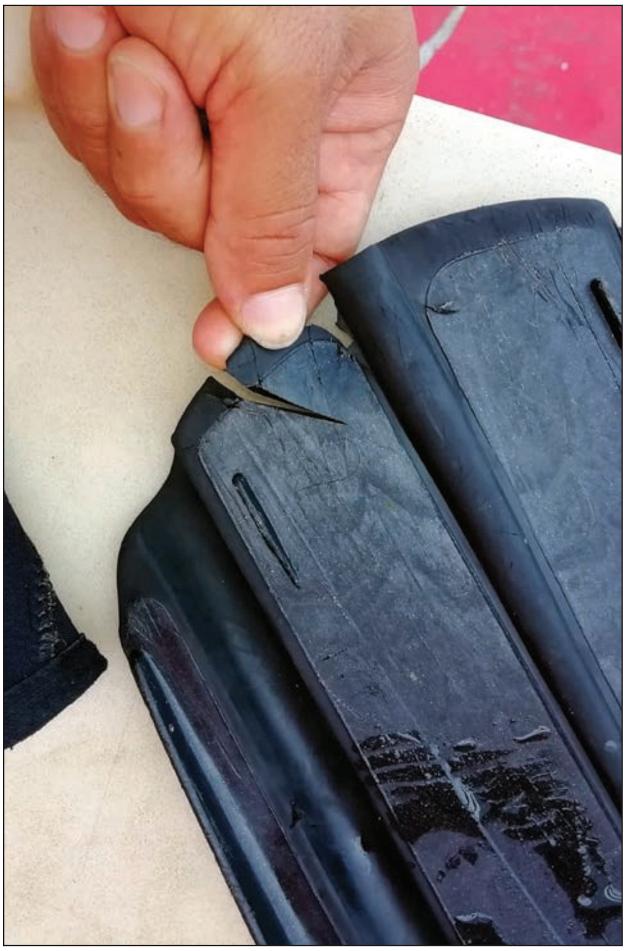


Fig. 3: Lacerations on fin belonging to diver 1 (Photo: İbrahim Yörüsün).
Sl. 3: Raztrganine na plavutki prvega potapljača (Foto: İbrahim Yörüsün).

circling around the diver 2 and suddenly one of the frenzied sharks attacked his fins from below (Fig. 4). Interview with the divers, as well as the detailed examination of the video footage was revealed that 7 to 8 specimens of sharks attacked the divers. Both attacks have resulted in deep lacerations on the dive gears (Figs. 3 & 4), and except of non severe injuries, no fatalities occurred. Divers 1 and 2 have ascended the surface as soon as possible, meanwhile they were trying to fend off the frenzied sharks with a spear gun. They were hauled out of the water with the aid of support vessel's crew; however, frenzied sharks continued to prey on dead or alive fishes for nearly one hour more.

The Sandbar shark, *C. plumbeus*, is one of the well-documented representatives of the Mediterranean carcharhinids (Serena, 2005; Saïdi *et al.*, 2005, 2007). Reproductive biology, and food and feeding habits of *C. plumbeus* were extensively investigated, based on the specimens captured in Gulf of Gabes (southern Tunisia, central Mediterranean; Saïdi *et al.*, 2005, 2007). It is a coastal-pelagic shark on continental and insular shelves and in deep water to maximum depth of 280 m (Serena, 2005). Its contemporary occurrence in Turkish Aeagean and Mediterranean coasts is also well-documented (see Kabasakal, 2019, for relevant references), and sandbar sharks are known regularly aggregated in Boncuk Cove, one of the best known nursery areas of *C. plumbeus* in the Mediterranean, between March and November (Filiz, 2018). During a two-year underwater video census survey, Filiz (2018) counted 275 mature sandbar sharks in Boncuk Cove, and no aggressive encounters with the sandbar sharks ever happened. Although, Boncuk Cove is a marine protected area and no human activities is allowed,

many tourism and fishing activities in the vicinity of the cove area are present; however, no attack to humans by sandbar sharks were recorded outside the cove region (H. Kabasakal, *pers. obs.*).

The occurrence of predatory sharks in the vicinity of marine aquaculture cages is well-documented in the Mediterranean Sea. Galaz and De Maddalena (2004) reported on a female white shark, *Carcharodon carcharias* (Linnaeus, 1758), with an estimated length of 5 m, tore the net of a 50 m diameter tuna cage, off Libya coast. Same authors also reported on blue sharks, *Prionace glauca* (Linnaeus, 1758), and shortfin mako sharks, *Isurus oxyrinchus* Rafinesque, 1810, trapped in tuna cages off Italian and Spanish coasts, respectively. Kabasakal (2014) reported on a white shark, which was attempting to tear the net of tuna cage, observed during the routine check of the condition of tunas by a diver. Although the shark has circled around the diver for a few times, fortunately, no attack occurred. Kabasakal & Gedikoğlu (2015) reported on a blue shark ($\geq$2 m total length), which was observed near aquaculture cages in Güllük Bay (southeastern Aegean Sea). In an extensive research on the site fidelity and movements of sharks associated with ocean-farming cages in Hawaii, Papastamatiou *et al.* (2010) observed that ocean fish cages appear to aggregate sandbar sharks. In the eastern Mediterranean, off Israeli coast, sandbar shark aggregations were also observed near power plants, where there is a continuous warm water outflow (Barash *et al.*, 2018).

C. plumbeus is a relatively large shark species armed with large, triangular teeth; however, this species has never been indicated to attack people, and is thought to be not particularly dangerous because of its strong preference for live fish (Compagno, 1984). Compagno (1984) summarized the aggressiveness of shark species in following three categories: (1) sharks that have attacked people or boats; (2) sharks suspected of attacking people; and (3) additional species of potential harmfulness, the last one which includes *C. plumbeus*, as well. Despite the Compagno's (1984) and Ebert & Stehmann's (2013) assessment that the sandbar shark is not particularly dangerous and has never been implicated on attacks to people, Caldicott *et al.* (2001) claimed that any shark that can grow larger than 1.8 to 2.0 m is potentially lethal to man. Ehrahardt *et al.* (1972) reported that sharks less than 2 m total lenght could be dangerous for humans. Their small size allow them to easily enter at lower depth in lagoons and attacks on divers or fishermen were recorded from French Polynesia. Fouques *et al.* (1972) reported an attack by a specimen of whitecheek shark *Carcharhinus dussumieri* (Müller & Henle, 1839), having 1.5 m total length. From this point of view, *C. plumbeus* can attain a maximum size of 3 m and common to 2.4 m TL (Serena, 2005; Ebert & Stehmann, 2013), thus, sandbar shark have enough potential to attack human, if provoked. From information collected from divers and fishermen Capapé *et al.* (1975) noted that the species could be considered as dangerous in Tunisian waters, as other carcharhinid species. Suspected shark of eight incidents listed in Global Shark Attack File (GSAF) is *C. plumbeus* (GSAF, 2020), of which the details are presented in table 1. Up to May 2020, 6523 shark attacks are logged in GSAF records.



Fig. 4: Lacerations on fin belonging to diver 2 (Photo: Özkan Karkın).
Sl. 4: Raztrganine na plavutki drugega potapljača (Foto: Özkan Karkın).

In conclusion, the present incidents were apparently provoked by the presence of excess amount of wounded and/or dead farmed fish, which caused a certain feeding frenzy of a shark species, normally not thought to be particularly dangerous. Fortunately, these provoked shark attacks have not ended with a

fatality; however, these incidents set forth the possibility of unexpected shifts in the behavior of a wild fish under stimulated conditions. A significant lesson learned from these incidents is that, as a precautionary measurement, aquaculture divers have been carrying out all diving operations related with routine clearance and maintenance of the fish farm, inside the cage, and no attack has been occurred since the date of the present incidents. *C. plumbeus* is a protected shark species in Turkish seas, and intentional or incidental captures, as well as landing of captured specimens, are strictly prohibited and any violations of the law, such as deliberately capturing and landing of a sandbar shark, would be imposed a cash fine. Aquaculture operations may have a potential to provide a significant and easy access source of additional forage to sharks. Since the ecological implications of the offshore or coastal cage farming are more complex to assess, future studies should be performed to determine the potential impacts of the cage farming on the foraging ecology of sandbar sharks, and other large predatory fish species with coastal occurrence in Turkish waters, as well.

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IZZVANI NAPADI SIVEGA MORSKEGA PSA, *CARCHARHINUS PLUMBEUS* (CARCHARHINIFORMES: CARCHARHINIDAE), OB OBALI TAŞUCU (SV SREDOZEMSKO MORJE, TURČIJA)

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POVZETEK

Šestindvajsetega avgusta 2019 je več sivih morskih psov (Carcharhinus plumbeus) *napadlo dva profesionalna potapljača na rutinskem potopu pri pregledovanju in čiščenju ribjih kletk. Sedem do osem morskih psov je sodelovalo v napadu na potapljača, ko sta odstranjevala v mrežo zapletene mrtve primerke rib na zunanji strani mreže. Globina, na kateri so ribje kletke zasidrane, je med 47 in 68 m, napad pa se je zgodil plitveje na globini okoli 20 m. Napade morskih psov je očitno sprožil prebitek ranjenih ali mrtvih gojenih rib in izzval skupinsko prehranjevalno razburjenost pri morskih psih, čeprav to vrsto ne smatrajo za agresivno.*

Ključne besede: sivi morski pes, Carcharhinidae, akvakultura, izzvan napad, ohranjanje, prehranjevalna razburjenost

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CAPTURE OF A LARGE GREAT WHITE SHARK, *CARCHARODON CARCHARIAS* (LAMNIDAE) FROM THE TUNISIAN COAST (CENTRAL MEDITERRANEAN SEA): A HISTORICAL AND ICHTHYOLOGICAL EVENT

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ABSTRACT

In this paper, the authors report an old capture of the great white shark, Carcharodon carcharias, *the first in the Gulf of Hammamet, central Tunisia. The specimen probably reached 6 metres in total length and weighed approximately two tons. Additionally, some comments about the distribution of the species in the area and throughout the Mediterranean are provided, emphasizing the presence of nursery grounds in this sea.*

Key words: Great white shark, size and total body weight, Gulf of Hammamet, *mattanza*, *Thunnus thynus*, nursery grounds

CATTURA DEL GRANDE SQUALO BIANCO, *CARCHARODON CARCHARIAS* (LAMNIDAE), LUNGO LA COSTA DELLA TUNISIA (MEDITERRANEO CENTRALE): EVENTO STORICO E ITTIOLOGICO

SINTESI

Gli autori riportano una vecchia cattura del grande squalo bianco, Carcharodon carcharias*, la prima nel Golfo di Hammamet, nella Tunisia centrale. L'esemplare probabilmente aveva raggiunto i 6 metri di lunghezza totale e circa due tonnellate di peso. Nell'articolo vengono inoltre forniti alcuni commenti sulla distribuzione delle specie nell'area in questione ed in tutto il Mediterraneo, sottolineando la presenza di zone di nursery nel bacino.*

Parole chiave: grande squalo bianco, dimensioni e peso corporeo totale, Golfo di Hammamet, *mattanza*, *Thunnus thynus*, zone di nursery

INTRODUCTION

Carcharodon carcharias (Linnaeus, 1758) is a large shark with worldwide distribution, especially in temperate waters, and its occurrence is well documented throughout the Mediterranean Sea following De Maddalena & Heim (2012). Most of these records occurred in the central Mediterranean, especially in the Strait of Sicily, where several juvenile and adult specimens were recorded (Quéro, 1984; Fergusson 1996, 2002; Saïdi *et al.*, 2005; Maliet *et al.*, 2013; Kabasakal, 2014). Off the Tunisian coast, *C. carcharias* is as well-known as other large and dangerous elasmobranch species (Capapé *et al.*, 1975). Bradaï & Saïdi (2013) noted that 59 reliable captures of *C. carcharias* were reported in the region between 1953 and 2012, the majority (56%) occurring in the Gulf of Gabès. Additionally, basic data collected through historical literature, previous published documents, interviews with fishermen, and personal observations suggested other occurrences of *C. carcharias* in Tunisian marine waters. Among them, a large specimen captured several decades ago and landed at the fishing site of Monastir (Fig. 1), which we are going to present.

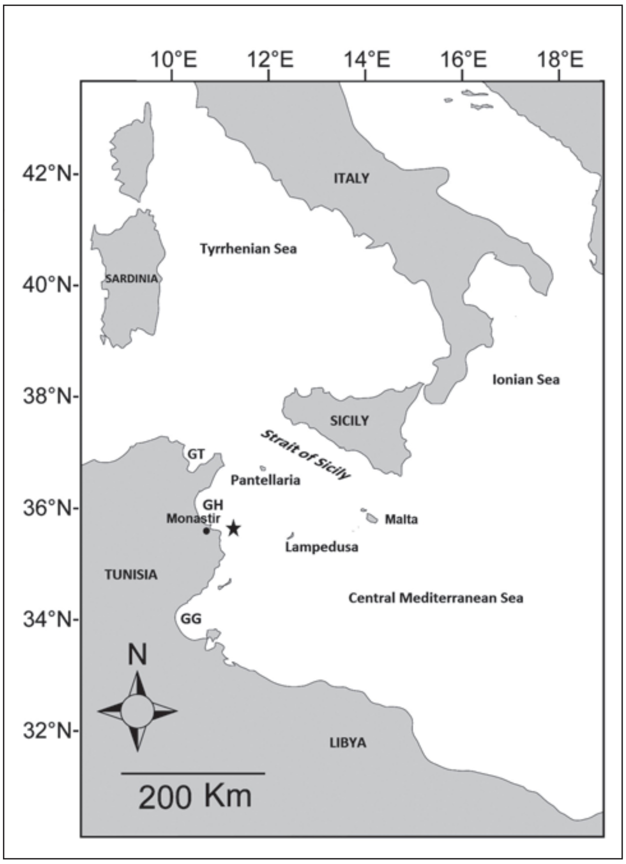


Fig. 1: Map of the central Mediterranean Sea, showing the capture site of the specimen of** Carcharodon carcharias **off the central Tunisian coast (black star). GT: Gulf of Tunis. GH: Gulf of Hammamet. GG: Gulf of Gabès.
Sl. 1: Zemljevid osrednjega Sredozemskega morja z označeno lokaliteto, kjer je bil ujet primerek vrste** Carcharodon carcharias **ob osrednji tunizijski obali (črna zvezdica). GT: Tuniški zaliv. GH: Hammameški zaliv. GG: Gabeški zaliv.

MATERIAL AND METHODS

The capture reported is that of a large female specimen of *C. carcharias* that occurred at the end of June 1975 in a *mattanza* set up in the waters surrounding the Kuriates Islands, located in the Gulf of Hammamet, central Tunisia (Fig. 1). A *mattanza* is an ancient traditional fishing technique used to catch thunnid species by trapping, mainly Atlantic bluefin tuna *Thunnus thynnus* (Linnaeus, 1758), when they are crossing the Mediterranean between February and July (Farrugio & Barbaroux, 2005). This kind of tuna fishing site is called a *tonnara* and is mainly targeted by large sharks, which can easily find available preys (Storai *et al.*, 2011).

Furtherly, the specimen of *C. carcharias* was cut into slices and rapidly sold by fishermen, its flesh being greatly appreciated in the local traditional cuisine. According to fishermen, the estimated total weight of these slices reached 2 tons. Only the jaws were removed and preserved (Fig. 2), and finally purchased for the personal collection of Mr Mohamed Zaouali, formerly Head of the Office National des Pêches of Tunisia, and late husband of JZ, one of the present co-authors.



Fig. 2: Jaws of the specimen of** Carcharodon carcharias **captured in the waters surrounding Kuriates Islands, central Tunisia, scale bar = 1000 mm (Photo: Hédi Zaouali).
Sl. 2: Žrelo belega morskega volka, ujetega v vodah okoli Kuriatskih otokov (osrednja Tunizija), merilo = 1000 mm (Foto: Hédi Zaouali).



Fig. 3: Specimen of *Carcharodon carcharias* ***landed at the fishing site of Monastir, observed by His Excellency, Habib Bourguiba, President of the Tunisian Republic, surrounded by members of his Government and other people. Habib Bourguiba was a lawyer and a nationalist leader who played a major role in leading his country to independence, earning the title of Supreme Combatant (Chadli, 2013). He governed Tunisia for three decades and, considering education as the highest priority, promoted the foundation and development of universities and specialized institutes (Belkhodja, 1999). The presence of President Habib Bourguiba at the landing site of Monastir is evidence of his genuine interest for all that concerned fisheries and science, which is clearly visible from the photo. The white arrow indicates an Atlantic bluefin tuna*** *Thunnus thynnus****, which was captured together with the great white shark (Photo: Mohamed Zaouali).***

Sl. 3: Primerek vrste *Carcharodon carcharias* ***si na ribji tržnici v Monastirju ogleduje Njegova Ekscelenca, Habib Bourguiba, predsednik Tunizijske Republike, obkrožen s člani njegove vlade in drugimi ljudmi. Habib Bourguiba je bil pravnik in vodja nacionalistov, ki je odigral ključno vlogo pri neodvisnosti Tunizije, zaradi česar so mu podelili naslov vrhovnega poveljnika (Chadli, 2013). Tunizijo je vodil tri desetletja, pri čemer je zagovarjal edukacijo kot glavno prioriteto ter promoviral ustanovitev in razvoj univerz in specializiranih inštitutov (Belkhodja, 1999). Njegova prisotnost v Monastirju kaže na njegovo zanimanje za ribištvo in znanost, kar potrjuje tudi fotografija. Z belo puščico je označen tun*** *Thunnus thynnus****, ki je bil ujet hkrati z morskim volkom (Foto: Mohamed Zaouali).***

The total length (TL) of the specimen was provided by the fishermen, however, to confirm this size, we have taken into consideration some methods recommended by Randall (1973) and Mollet *et al.* (1996). To estimate the size of a white shark, the latter authors used the relationships between the enamel height of the largest tooth in the upper jaw (UAE1) and the dried upper jaw perimeter (DUJP) *versus* total length; such relationships are detailed in Mollet *et al.* (1996, Tab. 1).

RESULTS AND DISCUSSION

The great white shark specimen was landed at the fishing port of Monastir together with *T. thynnus* in July 1975 (Fig. 3). It was a huge female measuring slightly under 6 m in total length, with the estimated total weight of the slices reaching 2 tons approximately following the accounts of fishermen who discovered some *T. thynnus* in its stomach contents. This was the first capture of *C. carcharias* in the Gulf of Hammamet and is probably the only capture known to date in this area. The height of the largest tooth of the upper jaw (UA1E2) was 60 mm, the DUJP 1200 mm. These measurements allow us to estimate the size of the Tunisian great white shark between 4.5 and 6 m total length following Mollet *et al.* (1996).

The records of largest *C. carcharias* captured to date concern two specimens, both reaching 7 m in total length. One specimen was caught by gill-net near Kangaroo Island, South Australia (Jury, 1987; Cappo, 1988) and designated as KANGA by Mollet *et al.* (1996), the other was caught by steel line baited by tuna and swordfish (Abela, 1989) in waters surrounding Malta Islands and designated as MALTA by Mollet *et al.* (1996). The DUJP in KANGA and MALTA were 1.250 m and 1.120 m, respectively, the UA1E2 51.6 mm and 46.9 mm, respectively (Mollet *et al.*, 1996). The total length was assessed between 5.1 and 7.3 m for MANGA, and between 4.5 and 6.4 m for MALTA (Mollet *et al.*, 1996). Following these results a total length of 6 m reported for the present Tunisian specimen remains a plausible hypothesis. The TL reported for MALTA suggested that its total body weight could be estimated between 2.4 and 3.6 tons. A great white shark captured in the Gulf of Gabès measured 5.87 TL and probably weighed more than 2 tons (Saïdi *et al.*, 2005). The estimated weight of the present *C. carcharias* – 2 tons – seems valid. Therefore, this specimen probably constitutes the largest great white shark known to date in Tunisian waters and one of the largest caught in the Mediterranean Sea. Comparatively, the largest Italian specimen of *C. carcharias* known to date, was estimated to reach a total length of 6.02 m, according to De Maddalena (1999).

Following Boldrocchi *et al.* (2017), *C. carcharias* exhibits a large range of prey species in stomach contents, indicating that it is a rather opportunistic feeder with a preference for scombrid and among them Thunna spp. This pattern explains why captures of great white shark generally occur close to both traditional and modern tuna fisheries (Boldrocchi *et al.*, 2017). However, it appears that the collapse of tuna fisheries enhanced the decline of these captures in some Mediterranean areas, such as Catalonian waters (Barrull & Mate, 2001), eastern Adriatic Sea (Soldo & Jardas, 2002) and both Sea of Marmara and the Bosphorus Strait (Kabasakal, 2016). Conversely, some authors have noted a possible relationship between *C. carcharias* and tuna farming (Boldrocchi *et al.*, 2017). The last *mattanza* site located off Sidi Daoud, north-eastern Tunisia, was closed in 2005 (Rhomdane *et al.*, 2014), and since then captures of white sharks were only reported locally (Bradaï & Saïdi, 2015). Some authors have noted a possible interrelationship between tuna and *C. carcharias*, due to the importance of thunnid species in the shark's diet (Boldrocchi, 2017).

A drastic decline of captures of *C. carcharias* in Tunisian waters remains uncertain and cannot be totally ruled out. However, based on the captures of pregnant females carrying developing embryos, young-of-the-year born in the wild and adults of both sexes, Fergusson (1996), Saïdi *et al.* (2005) and Bradaï & Saïdi (2013) suggested the presence of a nursery grounds for *C. carcharias* in the central Mediterranean Sea, even though further records are needed to confirm this opinion. Recent studies, such as Kabasakal (2014), Kabasakal & Gedikoğlu (2008), Kabasakal *et al.* (2018) indicated that *C. carcharias* currently occurs in the eastern Mediterranean Sea and in the Aegean Sea, as well as records of new-born and juvenile specimens. Similarly, a nursery ground for *C. carcharias* in north-eastern Aegean Sea remains a valid hypothesis.

Recruitment of *C. carcharias* in the Mediterranean Sea cannot be totally ruled out even if significant records are needed to confirm it. Intrusion of *C. carcharias* and other large elasmobranch species through the Suez Canal and the Strait of Gibraltar remains questionable (Capapé, 1989). Boldrocchi *et al.* (2017) noted that between 476 and 2015, 628 reliable records of *C. carcharias* were reported in the Mediterranean Sea, and informed us (Boldrocchi, 2020, *in letteris*) that no further record occurred in this region since 2015. Two records of *C. carcharias* reported by Rafrafi *et al.* (2015, 2019) from the north-eastern Tunisian coast are probably the last known to date in this sea.

ULOV VELIKEGA PRIMERKA BELEGA MORSKEGA VOLKA, *CARCHARODON CARCHARIAS* (LAMNIDAE) OB TUNIZIJSKI OBALI (OSREDNJE SREDOZEMSKO MORJE): ZGODOVINSKI IN IHTIOLOŠKI DOGODEK

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POVZETEK

Avtorji poročajo o starejšemu zapisu o pojavljanju belega morskega volka, Carcharodon carcharias*, ki je prvi podatek te vrste za Hammameški zaliv v osrednji Tuniziji. Primerek je verjetno v dolžino meril 6 m in tehtal približno dve toni. Nadalje avtorji razpravljajo o razširjenosti vrste na obravnavanem območju in širšem Sredozemskem morju ter o možnih razmnoževalnih okoljih te vrste (jaslicah).*

Ključne besede: beli morski volk, velikost in telesna teža, Hammameški zaliv, *mattanza*, *Thunnus thynus*, jaslice

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CAPTURES OF LARGE SHARK SPECIES FROM THE NORTHEASTERN TUNISIAN COAST (CENTRAL MEDITERRANEAN SEA)

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ABSTRACT

The paper examines three large predatory sharks captured in the north-eastern Tunisian coast and the central Mediterranean Sea. These three species are bluntnose sixgill shark Hexanchus griseus *(Bonnaterre, 1788), smalltooth sand tiger shark* Odontaspis ferox *(Risso, 1810), and great white shark* Carcharodon carcharias *(Linnaeus, 1758). The distribution of the three species is detailed and their ecological role commented.*

Key words: Chondrichthyes, *Hexanchus griseus*, *Odontaspis ferox*, *Carcharodon carcharias*, distribution, Tunisian coast

CATTURE DI GRANDI SPECIE DI SQUALI LUNGO LA COSTA TUNISINA NORD-ORIENTALE (MEDITERRANEO CENTRALE)

SINTESI

L'articolo esamina tre grandi squali predatori catturati lungo la costa nord-orientale della Tunisia e nel Mediterraneo centrale. Si tratta dello squalo capopiatto Hexanchus griseus *(Bonnaterre, 1788), del cagnaccio* Odontaspis ferox *(Risso, 1810) e del grande squalo bianco* Carcharodon carcharias *(Linnaeus, 1758). Gli autori presentano nel dettaglio la distribuzione delle tre specie e commentano il loro ruolo ecologico.*

Parole chiave: Chondrichthyes, *Hexanchus griseus*, *Odontaspis ferox*, *Carcharodon carcharias*, distribuzione, costa tunisina

INTRODUCTION

The northern Tunisian coast constitutes a transition path for fish species between the western and eastern Mediterranean Basins, and mainly for large predatory sharks (Quignard & Capapé, 1971; Capapé, 1989). This opinion has been corroborated by recent reports of captures occurring in this same area (Rafrafi-Nouira *et al.*, 2015, 2019; Ben Amor *et al.*, 2016; 2019; Capapé *et al.*, 2018).

Investigations regularly conducted concomitantly in Tunisian waters in the wake of local assistance of experienced fishermen have allowed the collection of a shoal of bluntnose sixgill shark *Hexanchus griseus* (Bonnaterre, 1788), a specimen of the rare smalltooth sand tiger *Odontaspis ferox* (Risso, 1810), and a great white shark *Carcharodon carcharias* (Linnaeus, 1758). This paper provides information and data about the capture of the three species, occurring during commercial surveys off the northern Tunisian coast, and some comments about the status of these species in the capture areas, and inside and outside the Mediterranean Sea.

MATERIAL AND METHODS

Each species is separately presented, including data of fishing gear, capture site, depth, nature of bottom and, when possible, associated fauna (Fig. 1). The specimens were rapidly cut into slices and sold soon after landing and it was generally difficult to get for each specimen its size, total length (TL) and total body weight. Photographs were taken to confirm the authenticity of these captures.

RESULTS

Bluntnose sixgill shark *Hexanchus griseus* (Bonnaterre, 1788)

This species is distributed worldwide, from the Pacific to the Indian Ocean, and off both sides of the Atlantic Ocean (Cook & Compagno, 2005). It is known throughout the Mediterranean Sea, in both eastern and western basins, and commonly collected in certain areas (Capapé *et al.*, 2003, 2004; Kabasakal, 2006, 2013; Basusta & Basusta, 2015).

Catches of *Hexanchus griseus* were previously cited in northern Tunisian areas at the level of Eskerkis Bank (Capapé, 1989; Rafrafi-Nouira *et al.*, 2015) and southwards, mainly in the Gulf of Gabès (Bradaï *et al.*, 2002). Additionally, single specimens of *H. griseus* were sporadically captured by trawl targeting European pilchard *Sardina pilchardus* (Walbaum, 1792), approximately at a depth of 200 m, in the Gulf of Tunis. On 10 June 2019, a shoal of 21 *H. griseus* was landed at the fishing port of Kélibia, located in the north of the Cape Bon Peninsula, northeastern Tunisia, by bottom longline targeting groupers. The captures occurred in the Strait of Sicily between Marettimo Island, close to the northwestern Sicilian coast, and the northeastern Tunisian coast, at a depth of 700-1000 m approximately (Ben Amor *et al.*, 2019).

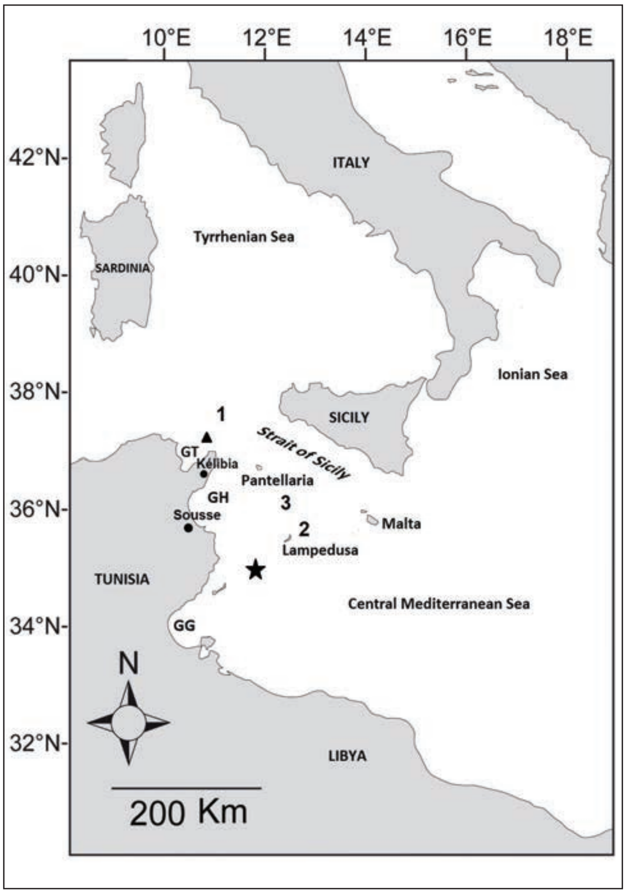


Fig. 1: Map of the central Mediterranean Sea, indicating capture sites of the three large sharks presented in this paper. *Hexanchus griseus****: 1. Capture from Ben Amor*** *et al.* ***(2019). 2. Capture occurring on 9 April 2020. 3. Capture occurring on 24 April 2020.*** *Odontaspis ferox****: black triangle, around Zembra Island from Capapé (1975). 2. Together with*** *Hexanchus griseus****. Black star, capture site of*** *Carcharodon carcharias****. GT = Gulf of Tunis. GH = Gulf of Hammamet. GG = Gulf of Gabès.***

Sl. 1: Zemljevid obravnavanega območja z označeno lokaliteto, kjer so bili ujeti primerki treh velikih morskih psov. *Hexanchus griseus****: 1. Podatki po Ben Amor s sod. (2019). 2. Ulov z dne 9 aprila 2020. 3. Ulov z dne 24 aprila 2020.*** *Odontaspis ferox****: črni trikotnik, okoli otoka Zembra, po Capapé (1975). 2. Skupaj s primerkom vrste*** *Hexanchus griseus****. Črna zvezdica označuje lokaliteto ulova vrste*** *Carcharodon carcharias****. GT = Tuniški zaliv. GH = Zaliv Hammamet. GG = Zaliv Gabès.***



Fig. 2: Shoal of* Hexanchus griseus *landed at the fishing site of Kélibia.
Sl. 2: Jata morskih psov šesteroškrgarjev, ujetih na lokaliteti Kélibia.

One year later, on 9 April 2020, a second shoal of 12 *H. griseus* was collected between the east coast of Linosa, an islet close to Lampedusa, and the western coast of Malta Island, at 35° 43′ 20″ N and 13° 9′ 43″ E (see Fig. 1). The specimens were caught by bottom longlines, at depths between 400 and 1100 m on rocky bottom, together with a specimen of smalltooth sand tiger shark *Odontaspis ferox* (Risso, 1810), a specimen of unidentified torpedo, several *Squalus blainvillei* (Risso, 1826), and a specimen of dusky grouper *Epinephelus marginatus* (Lowe, 1834). All captured specimens were landed at the fishing port of Kélibia, where they were examined and photographed (Fig. 2). The total weight of this shoal reached 55 tons.

Additionally, on 24 April 2020, other 2 specimens of *H. griseus* were captured off Pantelleria Island, at 36° 28′ 20″ N and 12° 31′ 30″ E. They were caught by bottom longlines, at a depth of 1100 m, on rocky bottom. They were not measured for length, but weighed 350 kg and 400 kg, respectively.

Finally, on 2 May 2020, 13 specimens of *H. griseus*, 2 females and 11 males, were captured 13 miles south-east of Pantelleria Island, at 36° 28′ 20″ N and 12° 29′ 51′ E. They were caught by bottom longlines, at a depth of 800 m on rocky bottom, together with specimens of *E. marginatus*. Their total body weight reached



Fig. 3: Fully yolked eggs removed from a female of* Hexanchus griseus*: 1. Each egg was covered by a fine diaphanous membrane. 2. All eggs were enveloped together in one single membranous capsule.
Sl. 3: Z rumenjakom bogato jajce, odstranjeno iz samice vrste* Hexanchus griseus*: 1. Vsako jajce je prekrito s fino presevno membrano. 2. Vsa jajca so skupaj združena v eni membranski kapsuli.

54 tons. Both females measured 3.5 m in total length and weighed 700 kg in total body weight, and carried 120 and 118 fully yolked eggs ready to be ovulated, respectively. Each egg was covered by a fine diaphanous membrane, and together they were enveloped in a single membranous capsule (Fig. 3).

All specimens of *H. griseus* were identified *in situ* or from photographs provided by Kélibia port authority, through the combination of some characteristics, such as: body stout, head broad, snout short and blunt, six gill slits, a single dorsal fin above fin base, upper jaw with 4 rows of front teeth, lower jaw with 6 rows of lower blade-like, comb-shaped teeth on each side, dorsal surface dark brown, belly beige. This description is in total accordance with Boeseman (1984), Compagno (1984), Quéro *et al.* (2003), and Ebert & Stehmann (2013).

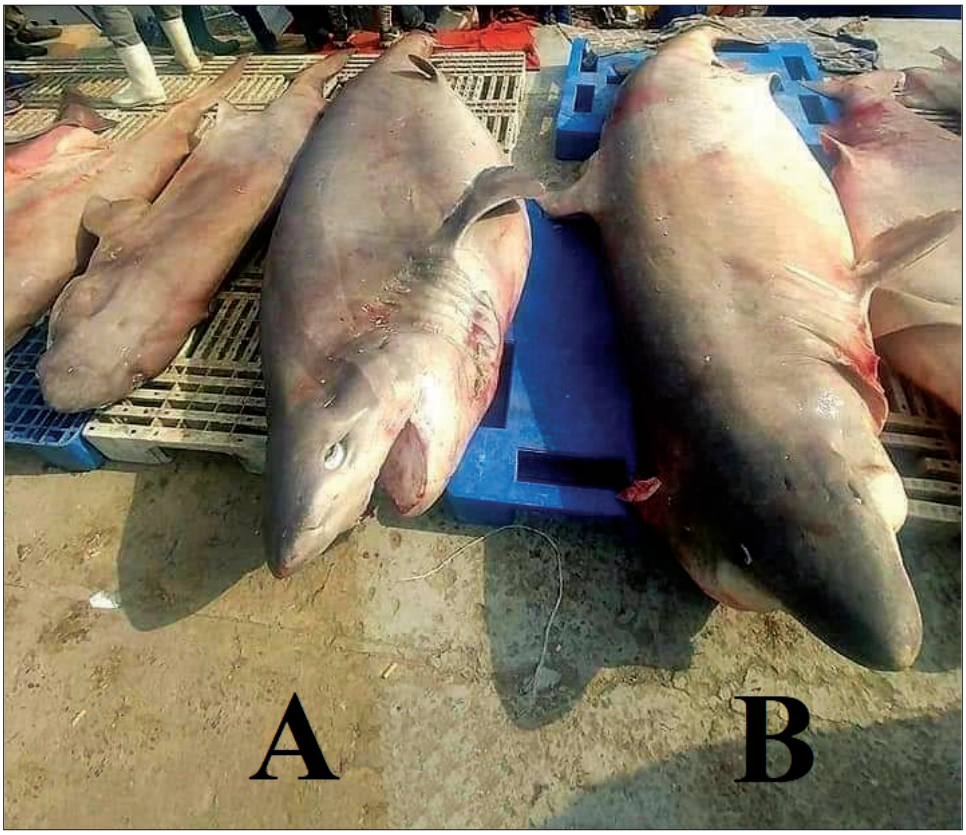


Fig. 4: A. *Hexanchus griseus*. ***B.*** *Odontaspis ferox*. ***landed at the fishing site of Kélibia.***
Sl. 4: A. *Hexanchus griseus*. ***B.*** *Odontaspis ferox*, ***iz ribiške lokalitete Kélibia.***

Smalltooth sand tiger shark *Odontaspis ferox* (Risso, 1810)

This shark was captured together with a shoal of *H. griseus* in similar conditions (see above). It was identified by the combination of the following main morphological characters: a bulky body with a long conical snout; eye small more than 4 times in snout, without nictitating eyelids; mouth long, extending behind the eyes (Fig. 4); teeth moderately large, each with a prominent narrow cusp and two or more pairs of lateral cusplets (Fig. 5); second dorsal fin origin above or slightly behind anal fin origin; anal fin and second dorsal fin smaller than first dorsal fin, caudal fin asymmetrical, with a strong lower lobe. Colour medium grey, sometimes with darker reddish spots scattered along the body, pelvic and anal fins very dark grey, lower edge of caudal fin almost black. These morphological characters are in total agreement with Compagno (1984). and Ebert & Stehmann (2013).

The smalltooth sand tiger shark, *Odontaspis ferox*, has a cosmopolitan distribution in warm temperate and tropical waters, and although essentially demersal, it has also been captured pelagically in mid-ocean (Compagno, 1984). The species often occurs inshore at steeply shelving coastal and insular locations, and in the Southern Hemisphere, most *O. ferox* were caught by trawl on the continental slope down to 880 m (Fergusson *et al*., 2008).

Due to its wide distribution, the species has been reported in oceans and seas throughout the world (Ebert & Stehmann, 2013). It is sporadically captured off the western Atlantic coast from North America (Sheehan, 1998) to Brazil (Menni *et al*., 1995). Off the eastern Atlantic coast, *O. ferox* is recorded off France (Quéro *et al*., 2003), Portugal (Carneiro *et al*., 2014) and southward of the Strait of Gibraltar from Morocco, without information about the capture site according to Collignon & Aloncle (1972), to Mauritania (Ebert & Stehmann, 2013). It is recorded in waters surrounding islands such as the Azores (Barcelos *et al*., 2018), Madeira (Maul, 1955), the Canaries (Barría *et al.,* 2018) and Cape Verde (Wirtz *et al*., 2013).

The first Mediterranean record of *O. ferox* occurred off Nice, southern France (Risso, 1810), subsequently, it was also reported by Bougis (1959) and Granier (1964); conversely, Capapé (1977) and Capapé *et al.* (2000) did not find the species in the area. Barrull & Mate (2002) considered the species as present off the Spanish coast. Tortonese (1956)

noted the capture of a large female, 370 cm TL, from the Gulf of Genoa, southwards, two other specimens were reported by Vanni (1992), while Vacchi & Serena (1997) and Sperone *et al.* (2012) reported captures from the Sicily Strait. Soldo & Jardas (2000) added that *O. ferox* is sporadically caught in the Adriatic Sea. Eastwards, *O. ferox* is present in the Aegean Sea (Ondrias, 1971), furtherly Kabasakal & Bayri (2019) summarized several captures of specimens that occurred in Turkish waters, while Akbora *et al.* (2019) reported other captures from Cyprus Island. Additionally, *O. ferox* is also recorded in the Levant Basin (Mouneimne, 1977; Golani, 2005; Bariche & Fricke, 2020).

Off the Maghreb shore, Dieuzeide *et al.* (1953) noted the occurrence of the species in the Algerian coast, which was furtherly confirmed (Hemida, pers. com., *in* Barrull & Mate, 2002). Capapé (1975) reported the capture of a female measuring 247 cm TL and weighing 70 kg, off Zembra Island, located in the Gulf of Tunis, northeastern Tunisia. The present capture (Fig. 5) constitutes a new record for the central Mediterranean Sea, where Schembri *et al.* (2003) noted that specimens may be collected annually in small numbers around Malta Islands; among them adult females of up to *ca* 360 cm TL were captured during years 1998 and 1999 in the area.



Fig. 5: Head and mouth of *Odontaspis ferox* ***showing the teeth (black arrow).***
Sl. 5: Glava in usta vrste *Odontaspis ferox* ***z zobmi (črna puščica).***

Great white shark *Carcharodon carcharias* (Linnaeus, 1758)

This large shark has worldwide distribution, especially in temperate waters, and its occurrence is well documented throughout the Mediterranean Sea, especially in the Strait of Sicily, where several juvenile and adult specimens were recorded (Quéro, 1984; Fergusson 1996, 2002; Saïdi *et al.*, 2005; Maliet *et al.*, 2013). *C. carcharias* is known off the Tunisian coast, like other large and dangerous elasmobranch species (Capapé *et al.*, 1975), and it appears that more than 60 reliable captures of the species should be taken into consideration from Tunisian waters (Zaouali *et al.*, 2020).

On 28 April 2020, a specimen was caught by drift longline baited with clupeid and scombrid species. The capture occurred off Sousse, a city located in eastern Tunisia, at 35° 01′ 01 N and 12° 11′ 11″ E, at a depth of 30-50 m, on soft bottom. The specimen was a female measuring 232 cm TL and weighing 90 kg. It was identified based on the combination of main morphological characters, such as: body very large, fusiform, snout rather blunt, flattened above, origin of first dorsal fin slightly posterior to inner corner of pectoral fin, a single keel on caudal base, teeth triangular, coarsely serrate, blade-like; colour greyish-brown or blue above, belly white (Fig. 6).

DISCUSSION

The captures of large sharks reported in the present paper indicate that these species are not totally extinct in the Mediterranean and some viable populations continue to develop and reproduce in this sea. Such is for instance the case for *H. griseus* and *C. carcharias*.

Conversely, such pattern is not evident for *O. ferox,* but it cannot be totally ruled out seeing that some specimens were recently captured in the eastern Mediterranean (Kabasakal & Bayri, 2019; Akbora *et al.*, 2019; Bariche & Fricke, 2020).

About the specimens caught from Malta Islands, Schembri *et al.* (2003) noted that such discoveries suggest that adult females reach annually selected sites, possibly for reproduction, which makes *O. ferox* especially vulnerable during aggregations when they fall prey to overfishing and spearfishing more easily. The recent captures of *O. ferox* from both central and eastern Mediterranean suggest that the species is not totally extinct in this sea. However, the presence of a viable population in some areas is yet uncertain, partially due to the lack of information about all traits of its reproductive biology.

These two new captures of shoals of *Hexanchus griseus* from the Tunisian coast confirm once again that the species is not facing a drastic decline despite its *K*-selective traits (Ebert, 1986; Capapé *et al.*,

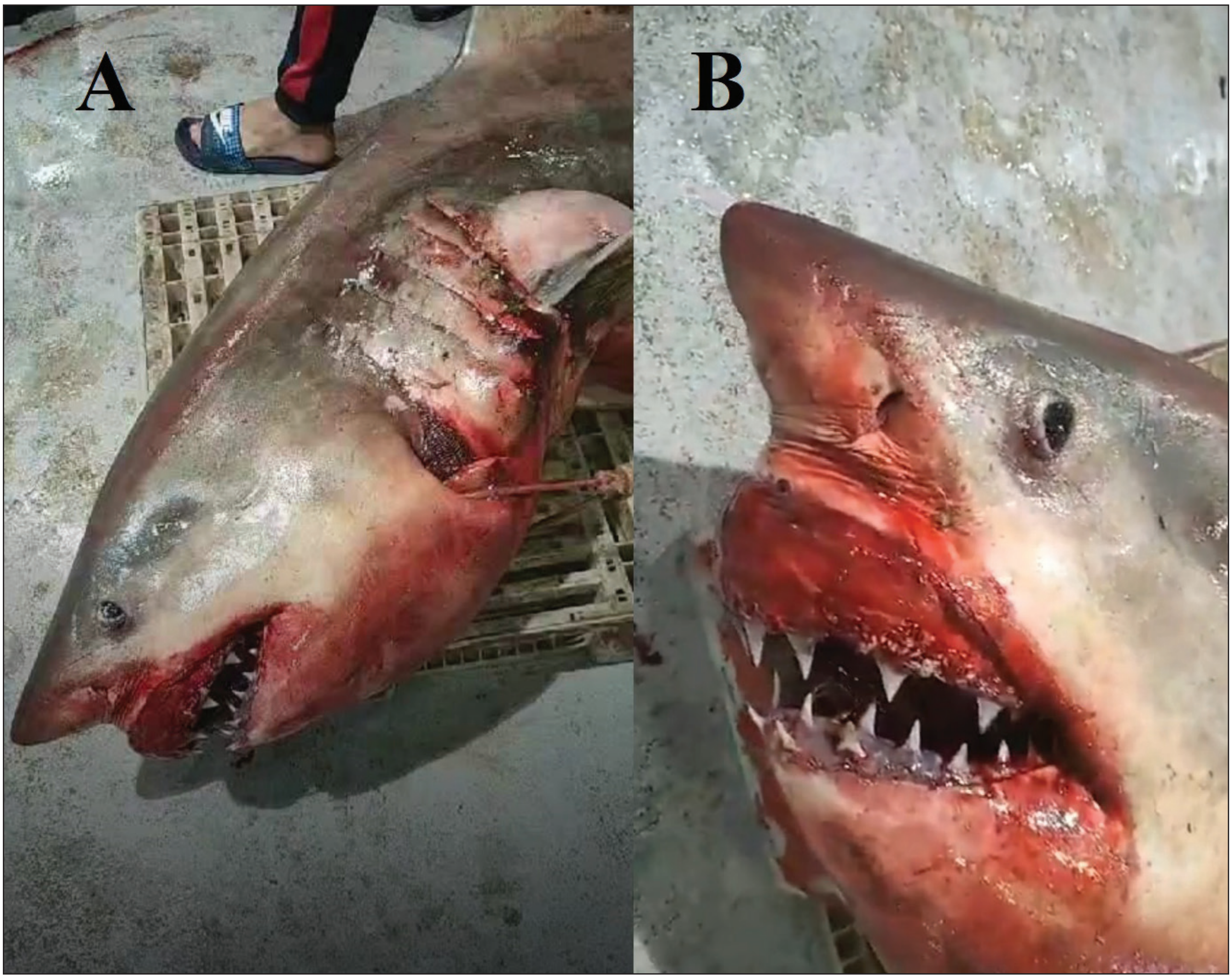


Fig. 6: *Carcharodon carcharias* ***landed at the fishing site of Sousse. A. Head. B. Snout showing teeth.***
Sl. 6: *Carcharodon carcharias* ***iz ribiške lokalitete Sousse. A. Glava. B. Gobec z zobmi.***

2004). Also, there have been several records reported throughout the Mediterranean during several decades, suggesting that a viable population of *H. griseus* is at present probably established in this sea. The captures of the two shoals probably occurred during the shark's reproductive period as indicated by the presence of large males and females, probably adults. Such hypothesis is confirmed by the fact that three females carried fully yolked oocytes. The record of first such specimen was provided by Ounifi-Ben Amor *et al.* (2017), and two are described in the present paper. The numbers of these oocytes, 85, 118 and 120 revealed higher levels of ovarian fecundity than is generally recorded in viviparous sharks. *H. griseus* could be classified as a relatively prolific elasmobranch species and such phenomenon explains its abundance in some areas, Tunisian and Turkish waters as best instances. Each oocyte is protected by a fine membrane, and all oocytes are enveloped together in an external membrane. This prevented them from being scattered while handled by fishermen, which unfortunately, can happen, too (see Ounifi-Ben Amor *et al.*, 2017).

Such abundance could be correlated to food sources and feeding aggregation, or, like in the present cases, to sharks apparently schooling to revisit sites with good environmental conditions and availability of food and in search of favourable nursery grounds where females could lay and protect their brood. Such patterns cannot be totally ruled out.

The flesh of *H. griseus* is not appreciated for consumption by Tunisian people, it is probably ichthyosarcotoxic due to its richness in oil and rather dangerous to health (see Capapé *et al.*, 1975). The shark was not targeted by fishermen, but due to the fact that the country is facing economic difficulties, these captures are considered an opportunity by fishermen and consequently the sharks are not released

back into the sea, which may contribute to a possible depletion of stocks. A strong monitoring of the species should therefore be conducted to avoid a drastic decline of the species in the area.

Boldrocchi *et al*. (2017) noted that between 476 and 2015, 628 reliable records of *C. carcharias* were reported in the Mediterranean Sea, and informed us (Boldrocchi, 2020, *in letteris*) that no other record occurred in this sea since 2015. The two records of *C. carcharias* reported by Rafrafi *et al*. (2015, 2019) from the northeastern Tunisian coast and the present record are probably the last ones known to date locally and in this sea. Such records mean that Tunisian waters are a hotspot for conservation and reproduction of this species. The specimen caught off Sousse is probably a juvenile specimen (see Compagno, 1984) and confirms the hypothesis that Tunisian waters could be a nursery ground for the species. Despite a number of white sharks caught in Tunisian waters and their relative abundance, their role in the conservation and reproduction of the species were completely dismissed by Moro *et al*. (2019), which is a rather incomprehensible and inadmissible opinion.

Following Ferretti *et al*. (2008) it appears, according to the evidence available, that large predatory sharks in the Mediterranean Sea are generally declining in abundance, diversity and range. Conversely, the data herein presented indicate that may not be the case in certain Mediterranean areas.

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ULOVI VELIKIH MORSKIH PSOV OB SEVEROVZHODNI TUNIZIJSKI OBALI (OSREDNJE SREDOZEMSKO MORJE)

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POVZETEK

Avtorji poročajo o treh vrstah velikih morskih psov, ujetih ob severovzhodni tunizijski obali. Gre za primerke morskega psa šesteroškrgarja Hexanchus griseus (Bonnaterre, 1788), morskega bika Odontaspis ferox (Risso, 1810), in belega morskega volka Carcharodon carcharias (Linnaeus, 1758). Razpravljajo o razširjenosti vrst in njihovi ekološki vlogi.

Ključne besede: Chondrichthyes, *Hexanchus griseus*, *Odontaspis ferox*, *Carcharodon carcharias*, razširjenost, tunizijska obala

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ON A HUGE SHORTFIN MAKO SHARK *ISURUS OXYRINCHUS* RAFINESQUE, 1810 (CHONDRICHTHYES: LAMNIDAE) OBSERVED AT CABRERA GRANDE, BALEARIC ISLANDS, SPAIN

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ABSTRACT

A huge female shortfin mako shark, Isurus oxyrinchus*, was observed on 28 June 2018 near Cabrera Grande, in the Balearic Islands, Spain. Its total length was carefully estimated at 500 cm based on a comparison with a 520-cm inflatable boat. This specimen is therefore the largest mako known alive and the second largest mako ever recorded worldwide.*

Key words: shortfin mako shark, *Isurus oxyrinchus*, maximum size, Spain, Cabrera National Park, Mediterranean Sea

IN MERITO A UN ENORME SQUALO MAKO DALLE PINNE CORTE *ISURUS OXYRINCHUS* RAFINESQUE, 1810 (CHONDRICHTHYES: LAMNIDAE) OSSERVATO ALLE ISOLE BALEARI, SPAGNA

SINTESI

Un'enorme femmina di squalo mako dalle pinne corte Isurus oxyrinchus*, fu osservato nei pressi di Cabrera Grande, alle Isole Baleari, in Spagna, il 28 giugno 2018. La lunghezza totale dell'esemplare fu stimata con accuratezza pari a 500 cm sulla base delle dimensioni di un gommone di 520 cm. Tale esemplare è pertanto il più grande mako mai osservato vivo ed il secondo più grande registrato a livello mondiale.*

Parole chiave: squalo mako dalle pinne corte, *Isurus oxyrinchus*, dimensioni massime, Spagna, Parco Nazionale dell'Arcipelago di Cabrera, mare Mediterraneo

INTRODUCTION

The shortfin mako *Isurus oxyrinchus* Rafinesque, 1810, inhabits temperate and tropical waters of the Atlantic, Pacific and Indian Oceans. It is pelagic, coastal and oceanic, occurring at a depth range from 0 to 500 m (Compagno, 2001). The shortfin mako is present in the entire Mediterranean (De Maddalena & Baensch, 2005), where it is caught mainly by tuna longline fishery and occasionally by swordfish fishery using longlines and driftnets (Celona *et al*., 2004; Megalofonou *et al*., 2005). Although the majority of shortfin mako catches are recorded in pelagic fisheries, in a recent report, Kabasakal (2015) emphasized that new-born and juvenile specimens of *I. oxyrinchus* can be incidentally captured by coastal stationary netting and bottom longline fishing, as well.

In the present article we report a record of a huge shortfin mako spotted in June 2018 by the team of Alnitak Marine Research and Education Center in the Balearic Islands, in Spanish Mediterranean waters.

MATERIAL AND METHODS

The observation took place from the research vessel Toftevaag, an 18-metre LOA converted historic Norwegian fishing boat, and from a 520-cm-long inflatable boat. The shark was encountered during a shipboard survey in the waters of the proposed extension of Cabrera National Park (Parque Nacional del Archipiélago de Cabrera), while tracking cetaceans, sea turtles, seabirds, devil rays and bluefin tuna. The crew of the Toftevaag at the time of the encounter was composed of Ricardo Sagarminaga van Buiten as captain, Fernando López-Mirones as biologist and filmmaker for ORCA-Films, and Beat von Niederhaeusern as boatswain. A few volunteers were also on board, including Sam Laederach, Georgina Stevens, Doris Juen, Cornelia Luxner, Susanne Luxner, Naim Lasgaa and Miguel Félix.

On 28 June 2018, at 07:00 UT, three hours after sunrise, at 39° 5.35 N and 003° 4.31 E, with wind force 0 (Beaufort) and sea state 0 (Douglas), the dorsal

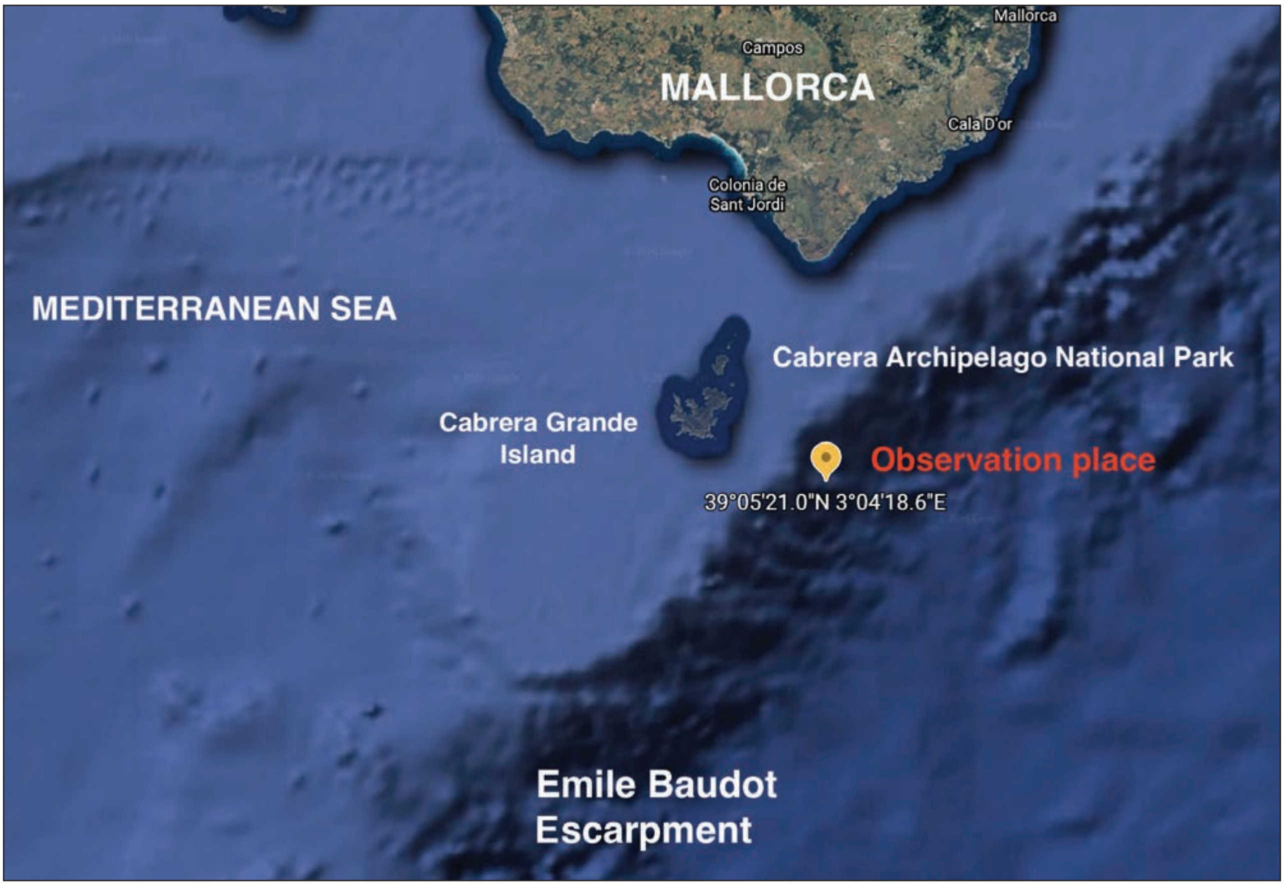


Fig. 1: Map showing the exact location where the estimated 500-cm TL female shortfin mako shark was observed on 28 June 2018.
Sl. 1: Zemljevid obravnavanega območja z označeno lokaliteto, kjer so avtorji 28. junija 2018 opazovali na 500 cm dolžine ocenjeno samico atlantskega maka.

fin was observed of a shark gliding slowly through the surface in 700-m-deep waters of the Emile Baudot escarpment, 5 nmi ESE of the Cabrera Grande Island (Fig. 1).

On the same day, a small juvenile loggerhead turtle *Caretta caretta* (Linnaeus, 1758) had been seen basking just one hour before 3.8 miles north from the sighting position, and a pod of 40 Risso's dolphins *Grampus griseus* (Cuvier, 1812) was observed slowly travelling north at a distance of 1000 m just after the encounter with the shark.

According to data from the Balearic Islands Coastal Ocean Observing and Forecasting System (SOCIB) and observation by Alnitak, 14 days prior to the shark encounter there had been particularly numerous observations of bluefin tuna *Thunnus thynnus* (Linnaeus, 1758), giant devil rays *Mobula mobular* (Bonnaterre, 1788), sperm whales *Physeter macrocephalus* Linnaeus, 1758 with calves, and large dolphin groups, while 7 days before the shark encounter there had been an important reduction in the number of sightings, except for turtles and Risso's dolphins, while sperm whales appeared to be silent, not clicking, just frequently breaching.



Fig. 2: The estimated 500-cm TL female shortfin mako shark, observed near Cabrera Grande, Balearic Islands, Spain, on 28 June 2018. This mako is believed to be the largest of its species observed alive, and the second largest ever recorded worldwide (photo by ALNITAK/ORCA-Films).
Sl. 2: Pet metrov dolga samica atlantskega maka, fotografirana 28. junija 2018 blizu otoka Cabrera na Balearih (Španija). Avtorji menijo, da gre za največji živeči primerek in drugi največji doslej zabeleženi primerek (foto: ALNITAK/ORCA-Films).

Pictures and videos of the shark were taken with a Nikon D750 camera, a Panasonic AG-DVX 200 video camera and a GoPro Hero3 video camera for subsequent analysis (Figs. 2-4). Underwater videos were filmed from the inflatable boat by the second author, but in them the shark was barely visible. A short, 25-second high-resolution aerial video was filmed by the first author from the mast of the boat from a height of 7 m, and was fundamental for identification purposes.



Fig. 3: Dorsal fin of the sighted shortfin mako shark (photo by ALNITAK/ORCA-Films).
Sl. 3: Hrbtna plavut opazovanega atlantskega maka (foto: ALNITAK/ORCA-Films).

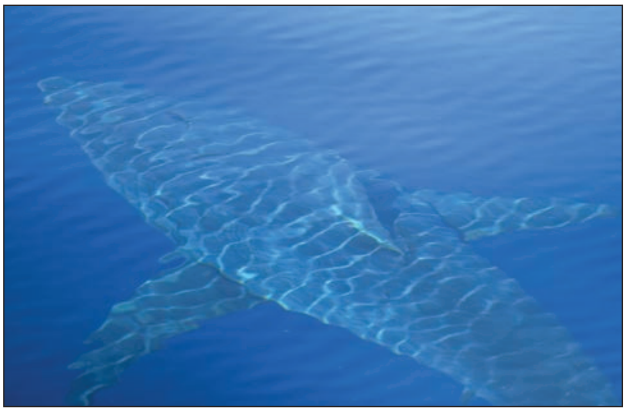


Fig. 4: Close-up of the shortfin mako shark showing the whitish band visible at the base of the dorsal fin, which is a peculiar feature of very large shortfin makos (photo by ALNITAK/ORCA-Films).
Sl. 4: Bližinski posnetek atlantskega maka, na katerem je razvidna belkasta proga na korenu hrbtne plavuti, ki je značilna za večje primerke te vrste (foto: ALNITAK/ORCA-Films).

RESULTS AND DISCUSSION

The shark was observed by the first two authors for 70 minutes. The size of the shark was carefully estimated by the first two authors at 500 cm total length (TL), based on the size of the inflatable boat, which was 520 cm LOA.

Some evident morphological features of the animal, including markedly spindle-shaped body, pointed conical snout, presence of caudal keel, and long gill slits, allowed the authors to make an immediate identification of the shark as a representative of the family Lamnidae. Although the shark was initially identified as a great white shark *Carcharodon carcharias* (Linnaeus, 1758), after careful examination of the high-resolution video and the photos we were able to conclude that the species observed by the team of Alnitak was actually an unusually large shortfin mako shark.

Large individuals of shortfin mako may bear a similarity with the great white shark, considerably more conspicuous than small or medium-sized individuals, however the two species can be told apart by various features (De Maddalena *et al.*, 2005). The identification of the Cabrera shark as *I. oxyrinchus* is therefore based on many morphological characteristics, which are listed below. Some of these also constitute a solid confirmation of the fact that the shark was indeed a shortfin mako of a very large size.

The estimated 500-cm size may seem barely acceptable for *I. oxyrinchus,* considering that only three other individuals of over 400 cm have been recorded to date worldwide (Kabasakal & De Maddalena, 2011), but on the other hand these records show that while the occurrence of such gigantic makos is extremely rare, it is still a reality.

The colouration is grey brown with a hint of blue. This can be observed in large makos, while smaller individuals display a much more brilliant blue colour with strong metallic reflection.

The shape of the first dorsal fin, high and more erect than in *C. carcharias*, corresponds to what is normally observed in *I. oxyrinchus* (Fig. 5).

The posterior margin of the first dorsal fin is mostly smooth and with only a few notches, which is typical of *I. oxyrinchus*, while *C. carcharias* shows a much more irregular posterior margin with a high number of notches (Fig. 5).

The whitish band visible at the base of the dorsal fin is a peculiar feature of very large shortfin makos and is not observed in great white sharks (Fig. 4).

The shape of the body, both the head and trunk stout and very massive, can suggest either a great white shark or a very large shortfin mako, but are definitely not compatible with small or medium-sized mako specimens.

The pectoral fins are long, conspicuously longer than in an average shortfin mako. It is known that neonate specimens of *I. oxyrinchus* have very short pectoral fins, however, these get conspicuously longer as the individual grows, up to the point of reaching a similar length as in *C. carcharias*.

The caudal fin's terminal lobe is not as prominent as in *C. carcharias*, and its size fits the one normally observable in *I. oxyrinchus* (Fig. 5).

The large size of the caudal keel may fit both *C. carcharias* and a very large *I. oxyrinchus*, but its angular shape definitely indicates the latter species.

The lower lobe of the caudal fin is as big as, or perhaps even bigger than, the upper lobe. In this regard we have to consider that while in *C. carcharias* the proportion of the two lobes tends to remain similar in small and large individuals, in *I. oxyrinchus* the lower lobe is much shorter than the upper lobe in juveniles, but becomes much larger when the individual grows bigger, and eventually it may match the size of the upper lobe.

On the head and the trunk there are bite scars that are likely the result of love bites by a mako, not by a great white shark.

The fast and somewhat nervous swimming pattern is typical of shortfin mako shark but not of great white shark.

Other features that differentiate *I. oxyrinchus* from *C. carcharias*, like the absence of a black tip on the underside of pectoral fins, the shape of the teeth, the respective position of the dorsal fin origin and pectoral fin free rear tip, the boundary between the colouration of the lateral and ventral surfaces, could not be observed. However, the totality of the observable features listed above, leads to a solid conclusive identification of the species as *I. oxyrinchus*.

While a study of 199 shortfin mako sharks showed an average total length of 171 cm (Kohler *et al.*, 1996) this species can sometimes attain incredibly larger sizes. The largest shortfin mako reported to date worldwide was a female caught in the late 1950s in the Aegean Sea off Marmaris, Turkey, which was estimated at 585 cm TL with a 577-619 cm range (Kabasakal & De Maddalena, 2011). Other large specimens have been recorded in the Mediterranean area. A 445-cm-long specimen was caught off Six-Fours les-Plages, France, in September 1973 (Capapé, 1977). A 425-cm-long shortfin mako was caught off La Galite Island, Tunisia, on 24 September 1876, and its jaws are preserved in the Natural History Museum of Genoa, Italy (Doria & Gestro, 1877). Lawley (1881) reported a 4-metre-long specimen that weighed 1000 kg, which was observed in a warehouse of a fishmonger in Livorno and was caught off Piombino, Italy. A 400-cm-long shortfin mako captured off Caska, Novalja, Croatia, on 13 May 1882 was reported by Brusina (1888). A 390-cm-long shortfin mako was caught on 30 November 1991 off Bagnara Calabra, Italy (Storai *et al.*, 2001). Another 390-cm-long specimen, weighing 513 kg, was caught on 20 September 2000 off Punta Alice, Italy (Storai *et al.*, 2001). A 390-cm-long female was caught on 26 July 2003 off Scaletta Zanclea, Italy, and another female, measuring 370 cm TL, was caught between Portopalo di Capo Passero and Marzamemi, Italy, on 22 June 2004 (Celona *et al.*, 2004). A 380-cm-long female was caught in summer 2012, by a commercial purse-seiner operating in İskenderun Bay, eastern Levantine Sea (Kabasakal, 2015).

Taking into account these records, the estimated 500-cm TL female shortfin mako shark observed near Cabrera Grande is believed to be the largest of its species observed alive, and the second largest ever recorded worldwide.

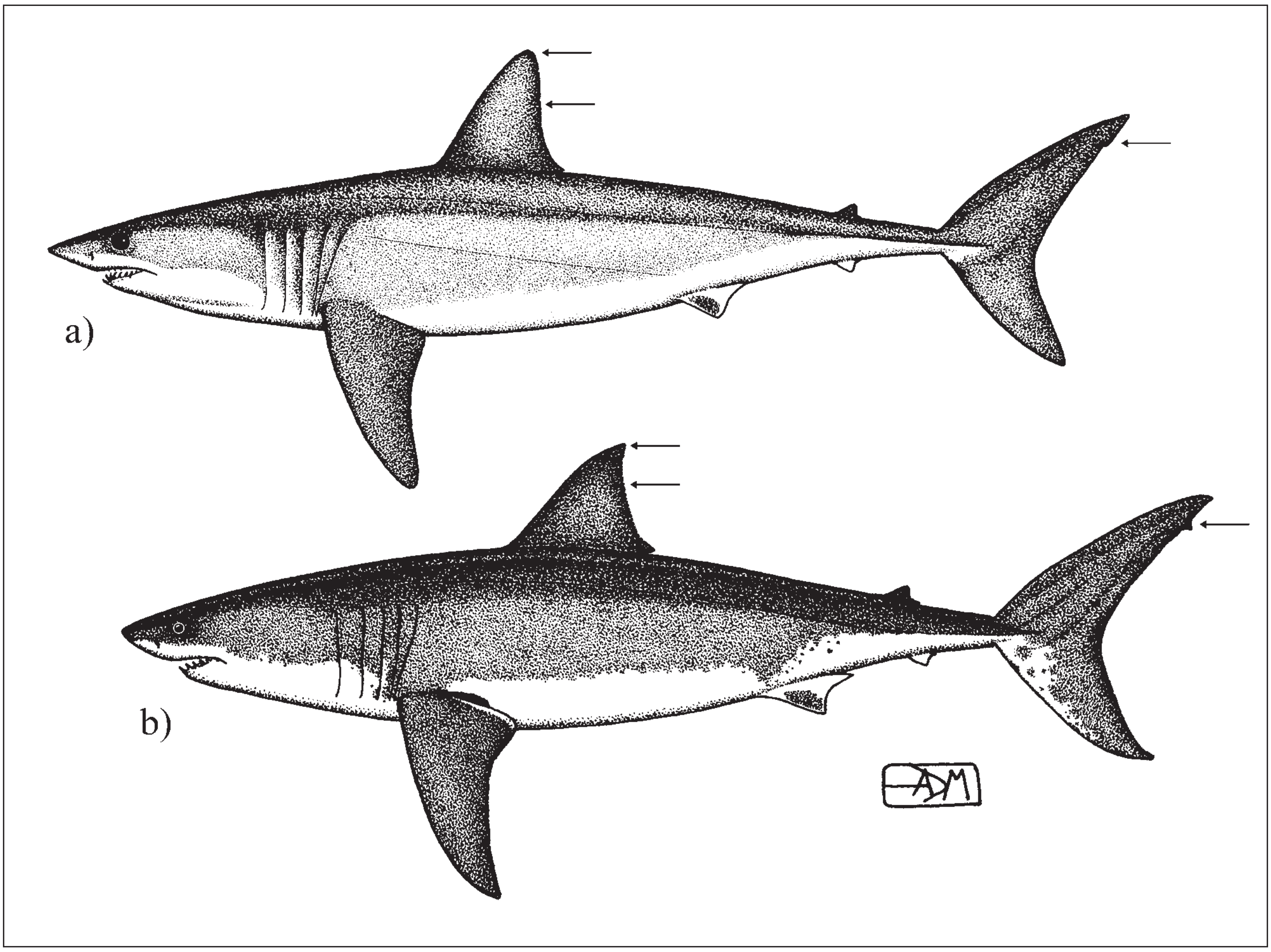


Fig. 5: Morphology of the shortfin mako (a) and the great white shark** Carcharodon carcharias **(Linnaeus, 1758) (b). The arrows mark some of the features that differentiate** I. oxyrinchus **from** C. carcharias**: a more erect first dorsal fin, a smoother posterior margin of the dorsal fin and a less prominent caudal fin's terminal lobe (illustration by Alessandro De Maddalena).

Sl. 5: Morfološke značilnosti atlantskega maka (a) in belega morskega volka** Carcharodon carcharias **(Linnaeus, 1758) (b). Puščice označujejo znake, po katerih je možno razlikovati vrsto** I. oxyrinchus **od** C. carcharias**: bolj pokončna hrbtna plavut, bolj gladek zadnji rob hrbtne plavuti in manj očitna krpica na repni plavuti (ilustracija: A. De Maddalena).

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O OPAZOVANJU VELIKEGA PRIMERKA ATLANTSKEGA MAKA *ISURUS OXYRINCHUS* RAFINESQUE, 1810 (CHONDRICHTHYES: LAMNIDAE) V BLIŽINI OTOKA CABRERA GRANDE, BALEARSKO OTOČJE, ŠPANIJA

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POVZETEK

*Osemindvajsetega junija 2018 so blizu otoka Cabrera Grande na Balearih (Španija) opazovali veliko samico atlantskega maka (*Isurus oxyrinchus*). Na podlagi primerjave z gumenjakom, ki je meril 520 cm, so ocenili, da je samica merila 500 cm v dolžino. Gre za največji živeči primerek te vrste in drugi največji doslej zabeleženi primerek.*

Ključne besede: atlantski mako, *Isurus oxyrinchus*, največja dolžina, Španija, Nacionalni park Cabrera, Sredozemsko morje

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CAPTURE OF A BIGEYE THRESHER SHARK *ALOPIAS SUPERCILIOSUS* (ALOPIIDAE) IN TURKISH WATERS (EASTERN MEDITERRANEAN SEA)

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ABSTRACT

The present paper reports a new capture of bigeye thresher shark, Alopias superciliosus *(Lowe, 1839). To date, 9 specimens have been recorded in the area, suggesting that a viable population of this type of shark might be successfully establishing in the area, but other records are needed to confirm this hypothesis. The paper comments on the distribution of the species in the Mediterranean Sea, suggesting that it does not originate from this sea, but is probably a migrant species from the eastern tropical Atlantic or from the Indian Ocean.*

Key words: Chondrichthyes, Alopiidae, distribution, population, migration

CATTURA DI SQUALO VOLPE OCCHIONE *ALOPIAS SUPERCILIOSUS* (ALOPIIDAE) IN ACQUE DELLA TURCHIA (MEDITERRANEO ORIENTALE)

SINTESI

Il presente documento riporta una nuova cattura di squalo volpe occhione, Alopias superciliosus *(Lowe, 1839). Ad oggi, nove esemplari sono stati registrati nell'area, suggerendo che una popolazione vitale di questa specie si stia stabilendo con successo nell'area. Saranno comunque necessarie altre segnalazioni per confermare questa ipotesi. L'articolo commenta la distribuzione delle specie nel Mediterraneo, suggerendo che lo squalo volpe occhione non sia originario di questo mare, ma sia probabilmente una specie migratrice arrivata all'Atlantico orientale o dall'Oceano Indiano.*

Parole chiave: Chondrichthyes, Alopiidae, distribuzione, popolazione, migrazione

INTRODUCTION

The bigeye thresher shark, *Alopias superciliosus* (Lowe, 1840), is a cosmopolitan species widely distributed in warm temperate waters of the the Atlantic, Pacific and Indian Oceans (Compagno, 1984). Off the eastern Atlantic coast, *A. superciliosus* is abundantly collected off the eastern side of the Atlantic Ocean from Portugal and Madeira to Morocco between 15° and 40° N (Quéro, 1984); Moreno & Morón (1992) and Fernandez-Carvalho *et al*. (2011) provided data about some traits of its reproductive biology from these areas.

A. superciliosus probably entered the Mediterranean Sea through the Strait of Gibraltar; in the Mediterranean, it was first recorded in the Ionian Sea following Gruber & Compagno (1981). An overview of Mediterranean records reported in literature shows that at least 40 specimens have been captured since, most of them in the eastern Basin, and especially in Turkish waters (Lanteri *et al*., 2017). Investigations regularly conducted in the latter area, focusing on elasmobranch species and supported by local fishermen actively helping the researchers, have enabled the collection of the specimen of *A. superciliosus* presented in this paper, which also provides comments about the species' origin and distribution in the same area and in the wider Mediterranean Sea.

MATERIAL AND METHODS

On 23 September 2012, a specimen of *A superciliosus* was captured by a pelagic longline at a depth of 1100 m off Fethiye, a city by the Aegean Sea, located at 36° 23′ N and 29° 00′ E (Fig. 1). The specimen was a female measuring 150 cm in total length (TL), and weighing 30.2 kg (Fig. 2). It was caught together with a Mediterranean moray eel *Muraena helena* Linnaeus, 1758, an oilfish *Ruvettus pretiosus* Cocco, 1829 and a swordfish *Xiphias gladius*, Linnaeus 1758.

RESULTS AND DISCUSSION

The specimen was identified *as A. superciliosus* based on the combination of the following main morphological characters: species-typical elongated upper lobe; snout rather long, bulbous; eyes very large, with orbits reaching the dorsal surface of the head; horizontal groove present on either side of head above the gills; labial furrows absent; first dorsal fin closer to pelvic fins than to pectoral fins; dorsal surface dark blue, belly cream to greyish.

These morphological characters are in total agreement with Compagno (1984), Quéro (1984) and Ebert & Stehmann (2013), and allow to include the present specimen among the nine *A. superciliosus* recorded to date in Turkish waters (Tab. 1). Ebert

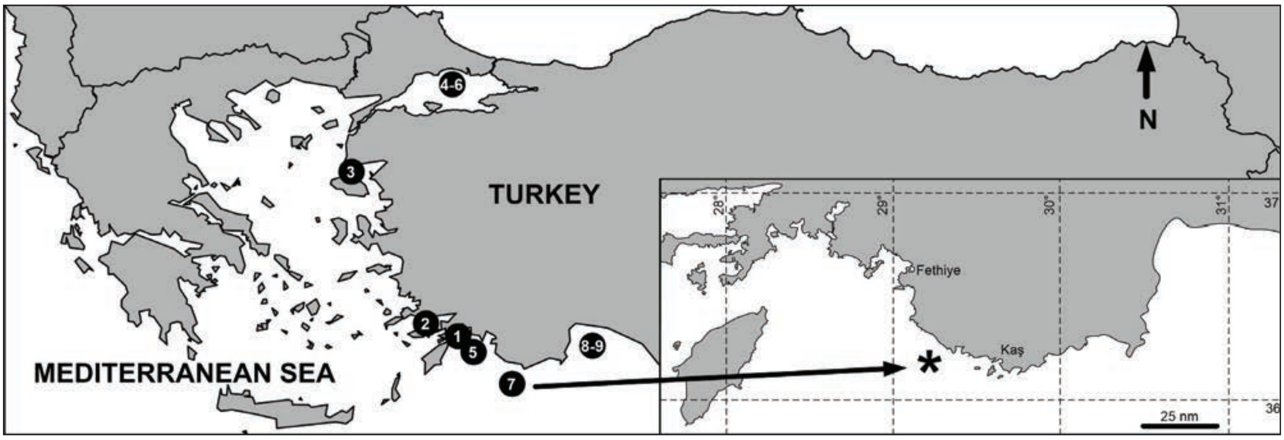


Fig. 1: Records of *Alopias superciliosus* ***captured in Turkish waters by chronological order. 1: Marmaris, Aegean Sea (Clo et al., 2009). 2: Gökova Bay, Aegean Sea (Kabasakal, unpubl. data). 3: Sivrice, Aegean Sea (Kabasakal et al., 2011). 4: Silivri, Sea of Marmara (Kabasakal & Karhan, 2007). 5: Fethiye, Aegean Sea (Kabasakal et al., 2011). 6: Silivri, Sea of Marmara (Kabasakal et al., 2011). 7: Fethiye, Aegean Sea (This study). 8-9: Antalya, NE Mediterranean (Soldo et al., 2014). Insert: asterisk indicates the capture site of the present specimen.***

Sl. 1: Zapisi o pojavljanju velikooke morske lisice v turških vodah po kronološkem redosledu. 1: Marmaris, Egejsko morje (Clo in sod., 2009). 2: Gökova Bay, Egejsko morje (Kabasakal, neobjavl. podatki). 3: Sivrice, Egejsko morje (Kabasakal in sod., 2011). 4: Silivri, Marmarsko morje (Kabasakal & Karhan, 2007). 5: Fethiye, Egejsko morje (Kabasakal in sod., 2011). 6: Silivri, Marmarsko morje (Kabasakal in sod., 2011). 7: Fethiye, Egejsko morje (pričujoča raziskava). 8-9: Antalya, SV Sredozemsko morje (Soldo in sod., 2014). Manjša slika - zvezdica označuje mesto ulova obravnavanega primerka.



Fig. 2: A - specimen of* Alopias superciliosus *captured off Fethiye (Aegean Sea), scale bar = 75 mm. B - head of the same specimen, scale bar = 75 mm.
Sl. 2: A - primerek velikooke morske lisice, ujet v vodah pred Fethiye (Egejsko morje), merilo = 75 mm. B - glava primerka, merilo = 75 mm.

& Stehmann (2013) noted that size at birth ranged between 100 and 140 cm TL, with the largest adult *A. superciliosus* ever captured reaching 484 cm TL. Since female specimens mature between 332 and 356 mm TL (Ebert & Stehmann, 2013), the present *A. superciliosus* could be considered at least as a juvenile female.

Serena (2005) noted that *A. superciliosus* was an occasional or rather rare species in the Mediterranean Sea. Conversely Clo *et al.* (2008) and Corsini-Foka & Sioulas (2009) considered the species abundant in some areas. De Maddalena & Baensch (2005) noted that the recent findings of *A. superciliosus* indicate that the shark occurs in the Mediterranean and is rather abundant in the eastern Basin, where a viable population appears to be progressively establishing (Kabasakal *et al.*, 2011). However, no nursery grounds for *A. superciliosus* were clearly observed anywhere in the Mediterranean Sea, and some traits of its reproductive biology are locally still unknown.

The status of the species in some Mediterranean areas remains questionable, due to fact that it is also noted as endangered (Walls & Soldo, 2016). Additionally, it is facing interspecific competition pressure from its closely sympatric species, thresh-

Tab. 1: Detailed records of specimens of* Alopias superciliosus *caught in Turkish waters.
Tab. 1: Podrobni zapisi o primerkih velikooke morske lisice, ujetih v turških vodah.

Record	Date	Area	Depth (m)	TL (cm)	Fishing gear	References
1	? /04/2005	Marmaris, Aegean Sea	?	?	?	Clo *et al.* (2009)
2	23/ 05/2005	Gökova Bay, Aegean Sea	?	350	?	Kabasakal (unpub. data)
3	21/05/2006	Sivrice, Aegean Sea	100	400	gill net	Kabasakal *et al.* (2011)
4	23/02/2007	Silivri, Sea of Marmara	?	450	purse-seine	Kabasakal & Karhan (2007)
5	28/02/2011	Fethiye, Aegean Sea	110	450	trammel net	Kabasakal *et al.* (2011)
6	02/07/2011	Silivri, Sea of Marmara	?	250	purse-seine	Kabasakal *et al.* (2011)
7	23/09/2012	Fethiye, Aegean Sea	1100	150	longline	This study
8	15/04/2015	Antalya, NE Mediterranean	500	?	?	Soldo *et al.* (2014)
9	05/05/2015	Antalya, NE Mediterranean	500	?	trawl	Soldo *et al.* (2014)

er shark *Alopias vulpinus* (Bonnaterre, 1788), which is locally somewhat more abundant. The best example of such competition is probably the Maghreb shore, where *A. superciliosus* does not occur and *A. vulpinus* is commonly caught (Hemida, 2005; Hemida 2019; *pers. comm.*, Rafrafi-Nouira *et al.*, 2019).

The first Mediterranean records of *A. superciliosus* occurred during 1952-1954 according to Corsini-Forkas & Sioulas (2009), with 40 other records reported since (Lanterni *et al.*, 2017). The first reports on the species came from the western Basin, suggesting a migration of *A. superciliosus* from the eastern tropical Atlantic to the Mediterranean Sea through the Strait of Gibraltar. It appears that most of the subsequent records of *A. superciliosus* reported by Lanterni et *al.* (2017) occurred in the eastern areas, therefore a migration of the species toward these areas remains a valid hypothesis.

A. superciliosus is known throughout the Indian Ocean, where viable populations are successfully established (Bass *et al.*, 1975; Das *et al.*, 2016). Although it is not recorded in the Red Sea (Golani & Fricke, 2005), a migration of the species into the Mediterranean Sea through the Suez Canal cannot be totally ruled out. It is evident that *A. superciliosus* does not originate from the Mediterranean Sea; rather, it is a vagrant species – a Herculean migrant (*sensu* Quignard & Tomasini, 2001) or a Lessepsian migrant (*sensu* Por, 1978), or having perhaps both origins. Similar patterns were also reported for the milk shark *Rhizoprionodon acutus* (Rüppell, 19837) by Ben Amor *et al.* (2016). Consequently, the origin of *A. superciliosus* in the Mediterranean could and should be determined using molecular tools. Still, whatever the origin of *A. superciliosus*, the latter should be defined as an alien species among the fish species known to date in this sea (see Golani *et al.*, 2017).

ULOV VELIKOOKE MORSKE LISICE *ALOPIAS SUPERCILIOSUS* (ALOPIIDAE) V TURŠKIH VODAH (VZHODNO SREDOZEMSKO MORJE)

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POVZETEK

Avtorji poročajo o novem primeru pojavljanja velikooke morske lisice, Alopias superciliosus *(Lowe, 1839). Do danes je bilo v turških vodah potrjenih 9 zapisov o pojavljanju te vrste, na podlagi katerih bi lahko sklepali, da se je viabilna populacija te vrste uspela ustaliti v obravnavanem območju, čeprav bi to hipotezo zanesljivo potrdili novi podatki. Avtorji razpravljajo o razširjenosti vrste v Sredozemskem morju, na podlagi katerih menijo, da vrsta ne domuje v njem, ampak je vanj najverjetneje zašla kot selivka iz vzhodnega tropskega Atlantika ali iz Indijskega oceana.*

Ključne vrste: Chondrichthyes, Alopiidae, razširjenost, populacija, selitev

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A RECORD OF RARE SPINY BUTTERFLY RAY, *GYMNURA ALTAVELA* (LINNAEUS, 1758), IN THE AMVRAKIKOS GULF (GREECE)

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ABSTRACT

During the study of the coastal fish assemblage of the Amvrakikos Gulf with underwater visual census methods a specimen of spiny butterfly ray (Gymnura altavela) *was sighted in the locality of Agios Georgios near Preveza on 12 June 2019. The specimen was observed and photographed on a sandy bottom at 9 m of depth.*

Key words: *Gymnura altavela*, critically endangered species, brackish environment, Greece

SEGNALAZIONE DI UNA SPECIE RAIFORME RARA, *GYMNURA ALTAVELA* (LINNAEUS, 1758), NEL GOLFO DI AMVRAKIKOS (GRECIA)

SINTESI

Durante lo studio della comunità ittica costiera del Golfo di Amvrakikos, con metodi di censimento visivo subacqueo, un esemplare di altavela (Gymnura altavela) *è stato avvistato nella località di Agios Georgios vicino a Preveza, il 12 giugno 2019. L'esemplare è stato osservato e fotografato su un fondale sabbioso a 9 m di profondità.*

Parole chiave*:* *Gymnura altavela*, specie in pericolo di estinzione, ambiente salmastro, Grecia

INTRODUCTION

The spiny butterfly ray, *Gymnura altavela* (Linnaeus, 1758), is a demersal batoid species present on both sides of the Atlantic Ocean (McEachran & Capapé, 1984). In the western Atlantic it occurs from southern New England to Brazil, in the eastern part from Portugal to Angola with the Canary Islands and Madeira included. It is also present in the Mediterranean and Black Seas, where it has been recorded more or less everywhere, but it is still considered a very rare species. It is known to inhabit shallow marine and brackish waters (Weigman, 2016). Due to its rarity in the Mediterranean Sea it is considered a critically endangered species (Abdul Malak *et al*., 2011). It was reported in the central Mediterranean Sea (El Kamel *et al*., 2009), in the Adriatic Sea (Dulčić *et al.,* 2003) and in many areas of the eastern Mediterranean Sea (see Özgür Özbek *et al*., 2016 and references therein). Recently, due to the findings of pregnant females of *G. altavela* carrying near-term embryos and small free-swimming specimens supposed to be neonates, Alkusairy *et al.* (2014) suggested that the area along the Syrian coast could be considered as a possible nursery area for *G. altavela*.

In this contribution we would like to share the information about the sighting of a specimen of spiny butterfly ray, *G. altavela*, observed in the Amvrakikos Gulf (western Greece) on 12 June 2019.

MATERIAL AND METHODS

The selected study area was the Amvrakikos Gulf, which extends over 405 km^2. It is a nearly enclosed gulf that maintains its connection with the Ionian Sea through a narrow channel. It is shallow, with a mean depth of 26 m and a maximum depth of 65 m (Rigas *et al.,* 2003). The embayment and its wetlands were proclaimed a National Park in 2008 (Zogaris & Dussling, 2010). The Amvrakikos Gulf is characterized by a eutrophic environment and near-brackish oceanographic conditions (Zogaris & Dussling, 2010). The coastal fish assemblage in the Amvrakikos Gulf was studied with the use of non-destructive underwater visual census methods. We used 25 m long and 5 m wide strip transects in three localities. A specimen of spiny butterfly ray was sighted on the transect in the locality of Agios Georgios near Preveza (20.802228 N; 38.957656 E) (Fig. 1) on 12 June 2019. It was observed on the sandy bottom at 9 m of depth. Photographs of the specimen were taken with a camera (Canon G7X Mk II) (Fig. 2).



Fig. 1: Map of the Amvrakikos Gulf (western Greece) with the locality where the specimen of *Gymnura altavela* ***was sighted.***
Sl. 1: Zemljevid zaliva Amvrakikos (zahodna Grčija) z označeno lokaliteto, kjer so opazovali primerek skata vrste *Gymnura altavela****.***

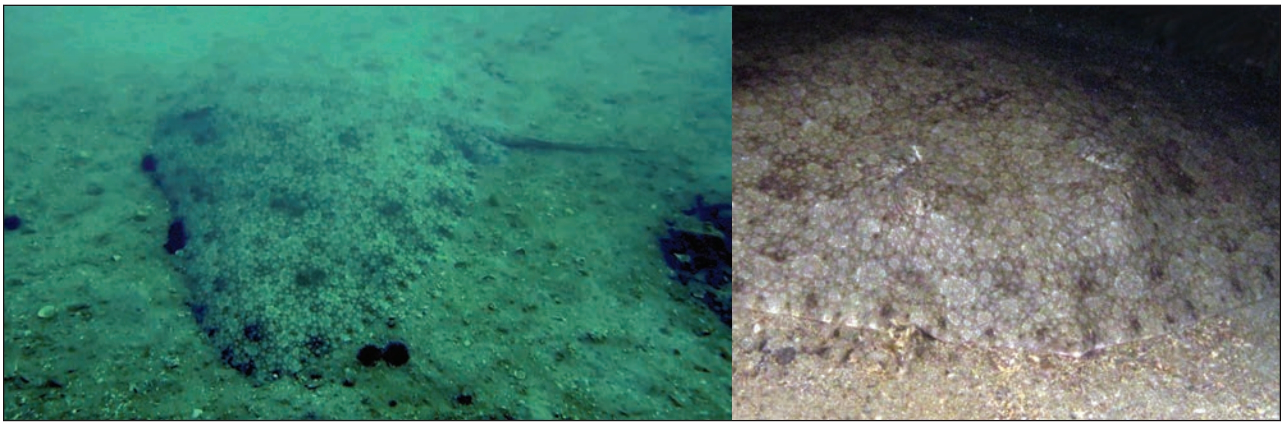


Fig. 2: The specimen of spiny butterfly ray photographed in the Amvrakikos Gulf, Greece, in June 2019. A – whole specimen, B – front view (Photo: S. Ciriaco).
Sl. 2: Primerek skata vrste** Gymnura altavela**, fotografiran 12. junija 2019 v zalivu Amvrakikos, Grčija. A – cel primerek, B – sprednji del primerka (Foto: S. Ciriaco).

RESULTS AND DISCUSSION

The specimen was easily recognized due to the peculiar shape of the disk, twice as wide as it is long. The tail is very short, only ¼ of the disk length, with one or two serrated spines. The snout is short and obtuse. The dorsal part is coloured with many dark and light spots, the ventral part is white. Juvenile and younger specimens have smooths skin (Bigelow & Schroeder, 1953). This is a large-sized batoid that can attain the maximum size of 1450 mm in disk width (Capapé, 1974).

The specimen of spiny butterfly ray was observed on a shallow sandy bottom. Although the visibility was low, we could estimate the disk width around 1100 mm. Also in other parts of the Mediterranean this species is associated with shallow areas (El Kamel *et al*., 2009). In the Gulf of Antalya, where this species is still present, the highest mean abundance and biomass values were recorded at a 25 m depth and decreasing towards deeper areas (Özgür Özbek *et al*., 2016). *Gymnura altavela* is known to be present in coastal and brackish areas, but also in euryhaline waters of lagoons (El Kamel *et al*., 2009) and highly eutrophic estuaries (Silva & Vianna, 2018). The area of Amvrakikos Gulf where the specimen was sighted is characterized by high eutrophication levels of pelagic compartments and a degradation of demersal ones, as already pointed out by Piroddi *et al*. (2016).

The specimen of *G. altavela* was found together with three large-sized specimens of the eagle ray, *Myliobatis aquila*. Other myliobatid rays had been observed in the Amvrakikos Gulf previously. Zogaris & Dussling (2010) reported on a sighting of twelve specimens of the bull ray (*Aetomylaeus bovinus*), two of them juvenile.

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ZAPIS O POJAVLJANJU REDKEGA SKATA VRSTE *GYMNURA ALTAVELA* (LINNAEUS, 1758), V ZALIVU AMVRAKIKOS (GRČIJA)

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POVZETEK

V okviru raziskav obrežne ribje združbe v zalivu Amvrakikos so z metodo podvodnih opazovalnih cenzusov 12. junija 2019 opazili primerek skata vrste Gymnura altavela *pri lokaliteti Agios Georgios blizu Preveze (Grčija). Primerek so opazovali, fotografirali in posneli na peščenem dnu na globini 9 m.*

Ključne besede: *Gymnura altavela*, kritično ogrožena vrsta, brakično okolje, Grčija

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SPAWNING PERIOD, SIZE AT FIRST SEXUAL MATURITY AND SEX RATIO OF THE ATLANTIC HORSE MACKEREL *TRACHURUS TRACHURUS* FROM BÉNI-SAF BAY (WESTERN COAST OF ALGERIA, SOUTHWESTERN MEDITERRANEAN SEA)

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ABSTRACT

Reproduction characteristics of the Atlantic horse mackerel, Trachurus trachurus *(Linnaeus, 1758), from Béni-Saf Bay were investigated. A total of 355 specimens were sampled between November 2015 and October 2017, comprising 47.04 % males, 44.79 % females and 8.17 % undetermined. The length of individuals ranged between 7.2 and 35.4 cm, and the weight from 5.28 to 312.7g. The length at first sexual maturity was evaluated at 15.6 cm for males and 14.9 cm for females. Variations in gonado-somatic index (GSI) showed that gonads of both sexes started to develop in late February and reached sexual maturity in May-June, which marks the spawning period of the species.* T. trachurus *from Béni-Saf Bay uses nutritional reserves mainly accumulated in spring to develop their sexual products for spawning in early summer.*

Key words: Atlantic horse mackerel, *Trachurus trachurus*, reproduction, Béni-Saf Bay, Algeria

PERIODO DI RIPRODUZIONE, DIMENSIONE ALLA PRIMA MATURITÀ SESSUALE E RAPPORTO TRA SESSI NEL SUGARELLO *TRACHURUS TRACHURUS* NELLA BAIA DI BÉNI-SAF (COSTA OCCIDENTALE ALGERINA, MEDITERRANEO SUD-OCCIDENTALE)

SINTESI

Gli autori hanno studiato le caratteristiche della riproduzione del sugarello, Trachurus trachurus *(Linnaeus, 1758), proveniente dalla baia di Béni-Saf. Un totale di 355 individui sono stati campionati tra novembre 2015 e ottobre 2017, con il 47,04 % di maschi, il 44,79 % di femmine e l'8,17 % di indeterminati. La lunghezza degli individui variava da 7,2 a 35,4 cm, e il peso da 5,28 a 312,7 g. La lunghezza alla prima maturità sessuale è stata valutata a 15,6 cm per i maschi e 14,9 cm per le femmine. Le variazioni dell'indice gonado-somatico (GSI) hanno mostrato che le gonadi di entrambi i sessi hanno iniziato a svilupparsi a fine febbraio e hanno raggiunto la maturità sessuale a maggio-giugno, il che evidenzia il periodo di riproduzione della specie. I sugarelli della baia di Béni-Saf utilizzano le riserve nutrizionali accumulate principalmente in primavera per sviluppare i loro organi sessuali per la deposizione delle uova all'inizio dell'estate.*

Parole chiave: sugarello, *Trachurus trachurus*, riproduzione, baia di Béni-Saf, Algeria

INTRODUCTION

The Atlantic horse mackerel, *Trachurus trachurus* (Linnaeus, 1758), is a gregarious species of the Carangidae family. It can be found in circa-littoral bottoms and even in the higher horizon of the bathyal zone (Athanassios & Konstantinos, 2015). This species is common in shallow coastal waters off the north-eastern Atlantic, from Iceland to the Islands of Cape Verde. It is also found in the Mediterranean, the sea of Marmara and more rarely in the Black Sea (Polonsky, 1969; Arneri, 1983), in the Eastern Channel and the North Sea. *T. trachurus* is a migratory species; it lives and hunts in shoals. Usually, it migrates towards the coasts in summer and returns to offshore waters in winter; it can be found close to the bottom where it can live between 50 and 400 m depth with a capacity to adapt to brackish water (Santic *et al.*, 2003). In the Mediterranean Basin *T. trachurus* is very common (Fezzani *et al.*, 2006), living in open water and near sandy bottoms; it feeds primarily on fish such as gobies, anchovy, sardine and only on certain shellfish (Ameri, 1983; Kerstan, 1985).

The study of the reproductive activity through the analysis of certain parameters such as variation of some biological indexes can help us better characterize the reproduction cycle by indicating the spawning period and the strategy of these fish. Several works have dealt with *T. trachurus* (Polonsky, 1969; Sedletskaya, 1971; Macer, 1974; Arneri, 1983; Arruda, 1984; Kerstan, 1985; Korichi,1988; Eaton,1989; Hecht,1990; Ben Salem & Ktari, 1994; Abaunza *et al.*, 1995; Kerstan,1995; Karlou-Riga & Economidis, 1996, 1997; Viette *et al.*,1997; Fezzani *et al.*, 2002; Abaunza *et al.*, 2003; Šantić *et al.*, 2008; Tahari, 2011; Aydin & Karadurmuş, 2012; Carbonara *et al.*,2012; Wahbi *et al.*, 2015; Aydin & Erdoğan, 2018; Gherram *et al.*, 2018; Azzouz *et al.*, 2019; Ferreri *et al.*, 2019). The present paper focuses on the reproductive biology of *T. trachurus* of Béni-Saf Bay, with an emphasis on the reproduction period and the size at first sexual maturity to complete gaps in the life cycle of this carangid species and to better manage this resource in this Mediterranean area.



Fig. 1: Map showing the exact location where the estimated 500-cm TL female shortfin mako shark was observed on 28 June 2018.
Sl. 1: Zemljevid obravnavanega območja z označeno lokaliteto, kjer so avtorji 28. junija 2018 opazovali na 500 cm dolžine ocenjeno samico atlantskega maka.

MATERIAL AND METHODS

A total of 355 specimens of *Trachurus trachurus* were collected from Béni-Saf fishery, fished by trawlers operating between 30-130 m depth, from November 2015 to October 2016 (Fig. 1). For each individual, total length (TL) was measured to the nearest millimetre, total weight (TW) and gonads weighed to the nearest 0.01 g. Fish lengths were classified in 1 cm group intervals (Fig. 2), and sex was determined macroscopically based on the morphology and the colour of the gonads (Mahdi *et al.*, 2018).

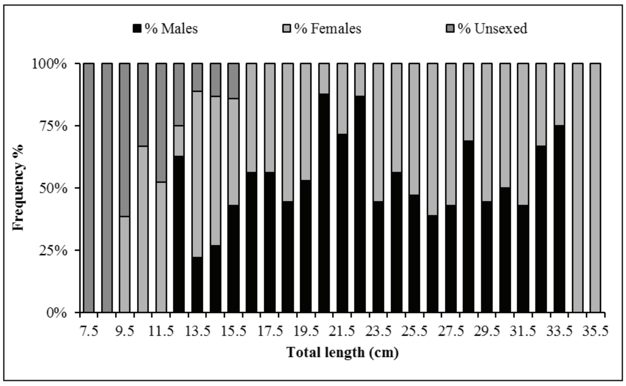


Fig. 2: *Trachurus trachurus* ***length frequency distribution of males and females caught in Béni-Saf Bay.***
Sl. 2: Porazdelitev dolžine pri samcih in samicah navadnih šnjurov, ujetih v zalivu Béni-Saf.

Sex ratio

Sex ratio is defined as being the proportion of the male or female individuals compared to the total number of individuals. It also gives an idea regarding the balance of the sexes within the population. The sex ratio generally relates the rate of femininity or masculinity of the population:

SR = F / (M + F) x 100
F= number of females,
M = number of males.

Gonado-somatic index (GSI)

In order to understand the sexual cycle and determine the spawning period the gonado-somatic index (GSI) was calculated monthly for both females and males, according to the equation below. The description of the reproductive cycle of this species and the determination of the spawning period were figured by tracking the monthly variations of this index.

$GSI = GW / TW \times 100$ (Ferreri *et al.*, 2019)

GW: gonads weight in g,
TW: total weight in g.

Coefficient of condition

The coefficient of condition K is defined by the relationship between the weight and the size of fish according to the equation:

$K = \frac{TW}{TL^3} \times 1000$ (Crim *et al.*, 1990; Ferreri *et al.*, 2019)

TW: total weight,
TL: total length.

Size at first sexual maturity

The size at first sexual maturity (Lm 50 %), which corresponds to the length at which 50 % of the individuals are mature, was calculated for our specimens. When considered ripe, the gonads occupy, or almost so, the totality of the visceral cavity. For males, the testes are milky white; for the females, the ovaries are bulky and pink, with oocytes visible through the ovary walls. For each size class (1 cm) previously defined, we counted mature individuals on one hand and immature individuals on the other. Consequently, we determined the relative proportions of each group in relation to the total size of each size class. We determined the values corresponding to the sizes at first maturity from an equation (1) whose curve is sigmoid:

$P = \frac{1}{1 + e^{-(b + aTL)}}$ (1) (Wahbi *et al.*, 2015) (1)

P: proportion of mature individuals,
TL: total length in cm,

The constants a and b are determined by the method of least squares transforming the equation (1) into linear type:

$Ln\left(\frac{p}{1 - p}\right) = b + aL$ (2)

RESULTS

Sex ratio

In total, 355 specimens of *Trachurus trachurus* were collected, 167 males (47.04 %), 159 females (44.79 %) and 29 unsexed (8.17 %). The length frequency distribution of the entire population is shown in (Fig. 2). Male length range was 12 to 33.5 cm; female length range was 8.8 to 35.4 cm. Male weight varied from 14.36 to 292.83 g and female weight varied from 5.28 to 312.78 g.

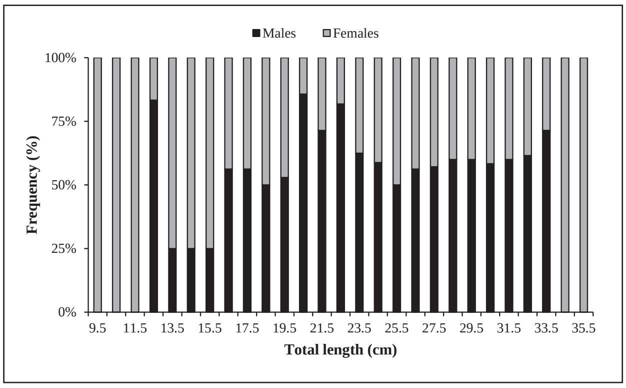


Fig. 3: Sex ratio of** Trachurus trachurus **by length size.
Sl. 3: Delež spolov pri primerkih navadnega šnjura glede na telesno dolžino.

Of the 355 individuals sampled sex ratio was in favour of males 1:0.95 and the $\chi 2$ test did not reveal any significant difference (p<0.05). In addition, the variations of sex ratio according to the size (Fig. 3), revealed by khi2 test significant differences in favour of females for length classes between 9.5 to 11 cm of TL ($\chi 2=11 > \chi 2_{t,0.05}=3.84$); beyond 16.5 cm of total length, males have the advantage but without significance (khi2). Beyond 34.5 cm of TL females are dominant.

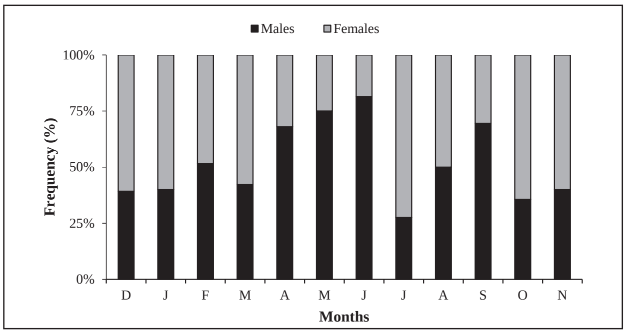


***Fig. 4: Monthly evolution of sex ratio of** Trachurus trachurus.*
Sl. 4: Delež spolov pri primerkih navadnega šnjura po mesecih.

Monthly variations of sex ratio (Fig. 4) reveal that females dominate during the months of November, October, December, January, March and July. Males outnumbered females during April, May, December, June and September, with numerical equality in August and February.

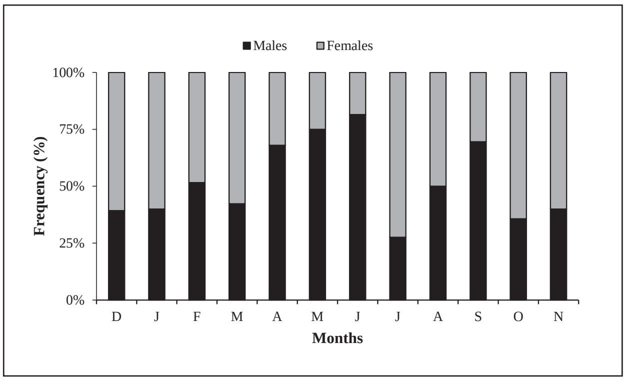


Fig. 4: Monthly evolution of sex ratio of *Trachurus trachurus***.**
Sl. 4: Delež spolov pri primerkih navadnega šnjura po mesecih.

Evolution of sex ratio related to seasons (Fig. 5) showed that females outnumbered males during the autumn-winter period, while males outnumbered females during the spring-summer period ($\chi2=5.54 > \chi2_{t,0.05}=3.84$) corresponding to the spawning period of *T. trachurus* in Béni-Saf Bay.

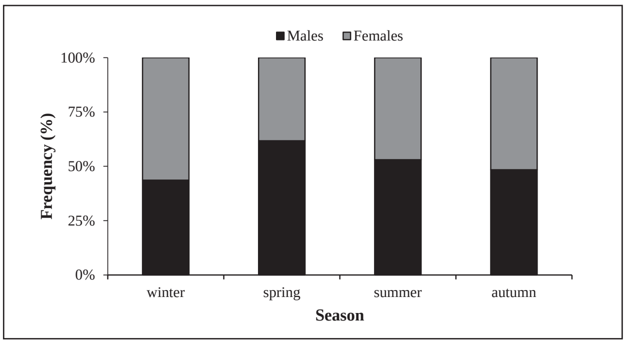


Fig. 5: Evolution of sex ratio of *Trachurus trachurus* **by seasons.**
Sl. 5: Delež spolov pri primerkih navadnega šnjura po sezonah.

Gonado-somatic index (GSI)¶

The monthly changes of the gonado-somatic index (GSI) allowed the determination of the spawning period during an annual cycle (Fig. 6); the monthly values of this index varied between 0.47 and 5.69 for females and between 0.37 and 4.98 for males. From February, this index increases for both sexes to attain a maximum in June, after which values begin to gradually decrease until they reach their lowest value in October for males and females.

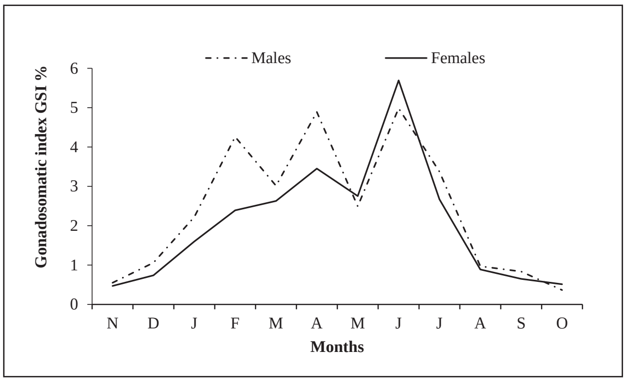


Fig. 6: Monthly evolution of gonado-somatic index for males and females of *Trachurus trachurus***.**
Sl. 6: Gonadosomatčni indeks pri samicah navadnega šnjura po mesecih.

Gonado-somatic index related to size classes

To investigate the role of small specimens and their contribution to the renewal of the resource we linked GSI to the total length of individuals (Fig. 7) and it was established that GSI increased simultaneously with length.

For males, we recorded a maximum GSI at 5.94, corresponding to a size 26.5 cm TL, and a minimum of GSI with 12.5 cm TL. For females, the GSI maximum value was recorded at 4.89 corresponding to a size of 22.5 cm TL, while the GSI minimum was recorded for a size of 13.5 cm TL.

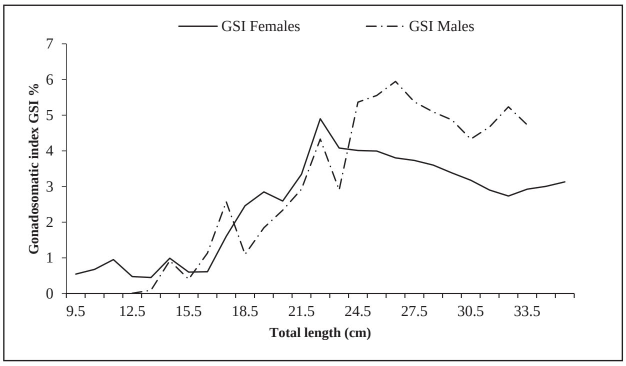


Fig. 7: Evolution of gonado-somatic index related to total length of males and females of *Trachurus trachurus***.**
Sl. 7: Gonadosomatčni indeks pri navadnem šnjuru glede na celotno dolžino samcev in samic.

Condition factor K

Evolution of the condition factor coefficient K seems to be closely related to the gonado-somatic ratio GSI, but the two indices tended to be inversely proportional. In fact, during the spawning period, the condition factor of the specimens is recorded at its lowest values (7.05) and in rest period the same factor records its highest values (8.54). The maturation of the sexual products and their emissions requires relatively high energy expenditure, and as a consequence the fish weight during spawning period is reduced (Figs. 8 and 9), corresponding to a reduction in condition factor of ~14 and 20 % for males and females, respectively.

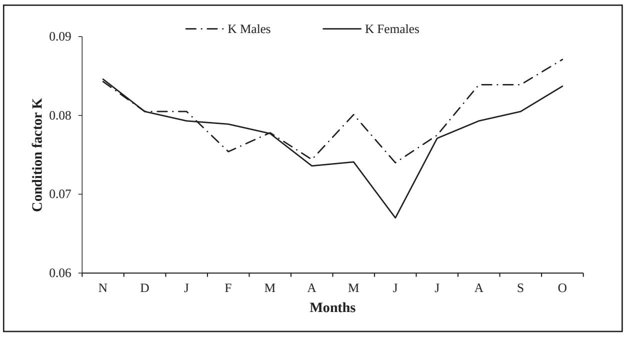


Fig. 8: Annual evolution of condition factor K of** Trachurus trachurus **males and females.
Sl. 8: Letna dinamika kondicijskega faktorja K pri samcih in samicah navadnega šnjura.

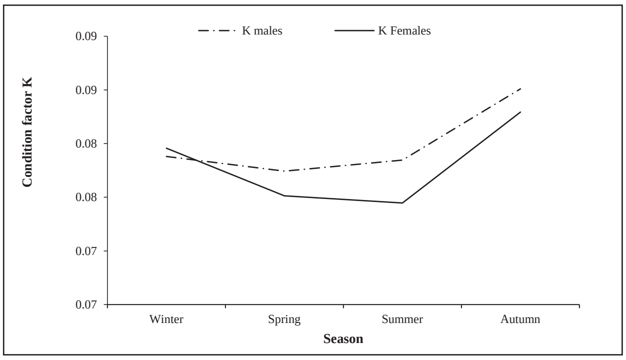


Fig. 9: Seasonal evolution of condition factor K of** Trachurus trachurus **males and females.
Sl. 9: Sezonska dinamika kondicijskega faktorja K pri samcih in samicah pri primerkih navadnega šnjura.

Size at first sexual maturity

The proportion of mature individuals in each size class (Fig. 10) showed that first maturity was attained at 14.9 cm TL where 50 % of the females were mature, whereas 50% of males attained this proportion at 15.6 cm TL.

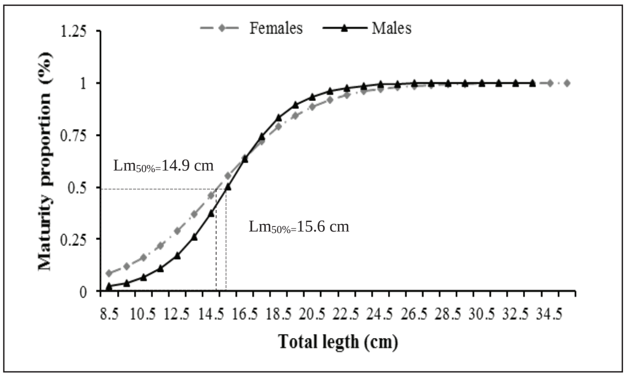


***Fig. 10: Length at sexual maturity of males and females of** Trachurus trachurus.*
Sl. 10: Dolžina, pri kateri samci in samice navadnega šnjura dosežejo spolno zrelost.

DISCUSSION

The sex ratio is slightly in favour of the males. The evolution of this index does not have phrenological regularity and is close to 1 for the March-June period, whereas females dominate in July. The Atlantic mackerel is a pelagic fish living in dense fish benches. It is possible that certain fish populations are predominantly males or females. According to Carbonara *et al.* (2012) and Wahbi *et al.* (2015) fluctuations of the sex ratio are due to ethologic phenomena (stray species, demographic segregations) responsible for the overdispersion and segregated distribution of the sexes. The difficulty in interpreting the fluctuations of this ratio is due to several factors, such as the behaviour of the species, the spawning period and mortality, sampling procedure, aggregation of the of the same sex individuals, etc. The change in weight of the ovaries and the testes during the cycle of maturation shows that the gonads develop at stage II, increased at stage III then regress at stage IV. The weight of the testes is higher than that of the ovaries during this last phase; this could indicate intense expulsion of the sexual products of females.

The monthly evolution of GSI follows a similar pattern for the two sexes, the spawning starting at the beginning of February to continue until July. The period of reproduction (Tab. 1) extends from February to July with a peak in June, while in central Algerian waters (Bousmail Bay), the reproduction is during summer and at its maximum in July-August. In ccertain areas (Spain, Portugal, France and Morocco) *T. trachurus* has an early spawning period beginning in spring with a maximum around March and all

Tab. 1: The spawning period and length at first maturity obtained for* Trachurus trachurus *by various authors.
Tab. 1: Obdobje drstitve in dolžina, ko šnjur* Trachurus trachurus *doseže spolno zrelost po različnih avtorjih.

<table>
<tr><th>Authors</th><th colspan="2">Area</th><th>Lm50% (cm)</th><th>Spawning period</th></tr>
<tr><td>Polonsky, 1969</td><td colspan="2">North Sea and English Channel</td><td>20 - 24 *</td><td>-</td></tr>
<tr><td>Sedletskaya, 1971</td><td colspan="2">North Africa</td><td>16 - 23</td><td>-</td></tr>
<tr><td>Macer, 1974</td><td colspan="2">North Sea and English Channel</td><td>20 - 24</td><td>March to August</td></tr>
<tr><td>Arneri, 1983</td><td colspan="2">Adriatic</td><td>15 - 18</td><td>-</td></tr>
<tr><td rowspan="3">Arruda, 1984</td><td rowspan="3">Portuguese coast</td><td>Western coast</td><td rowspan="3">16 - 19 M
21 - 24 F</td><td>November to May</td></tr>
<tr><td>Matosinhos Bay</td><td>April to December</td></tr>
<tr><td>Southern coast</td><td>Whole year</td></tr>
<tr><td>Kerstan, 1985</td><td colspan="2">North -East Atlantic (British water)</td><td>24.2 - 24.6</td><td>-</td></tr>
<tr><td>Korichi, 1988</td><td colspan="2">Algiers Bay (Bou-Ismail bay)</td><td>14.2 *</td><td>-</td></tr>
<tr><td>Eaton, 1989</td><td colspan="2">West of the British Isles</td><td></td><td>March to July</td></tr>
<tr><td>Hecht, 1990</td><td colspan="2">South-East coast of South Africa</td><td>32 - 33</td><td>June to November</td></tr>
<tr><td>Abaunza et al., 1995</td><td colspan="2">North-west of Spain
Galician and Cantabrian shelf</td><td>20.9 M
21.9 F</td><td>February to May</td></tr>
<tr><td rowspan="3">Kerstan,1 995</td><td colspan="2">Southwest coast of Irland</td><td>19.8 M
25 F</td><td>-</td></tr>
<tr><td colspan="2">Northern biscary</td><td>19.4 M
24.6 F</td><td>-</td></tr>
<tr><td colspan="2">South of biscary</td><td>19 M
25.3 F</td><td>-</td></tr>
<tr><td>Karlou-Riga & Economidis, 1996</td><td colspan="2">Aegean Sea</td><td>22</td><td>-</td></tr>
<tr><td>Karlou-Riga & Economidis, 1997</td><td colspan="2">Saronikos Gulf (Greece)</td><td>-</td><td>December to April</td></tr>
<tr><td>Viette et al.,1997</td><td colspan="2">Italy Gulf of Trieste</td><td>15.6 M
16 F</td><td>May to August</td></tr>
<tr><td>Abaunza et al., 2003</td><td colspan="2">Northwest Atlantic</td><td>16 - 25</td><td>February to August</td></tr>
<tr><td>Šantić et al., 2008</td><td colspan="2">Eastern Adriatic Sea</td><td>-</td><td>December to May</td></tr>
<tr><td>Tahari, 2011</td><td colspan="2">Oran Bay (Algeria)</td><td>-</td><td>October to March</td></tr>
<tr><td>Aydin & Karadurmuş, 2012</td><td colspan="2">Ordu Black Sea (Turkey)</td><td>-</td><td>May to August</td></tr>
<tr><td rowspan="3">Carbonara et al., 2012</td><td colspan="2" rowspan="3">Central-Western Mediterranean Sea</td><td>GSA 10
17.8 M
18.9 F</td><td rowspan="3"></td></tr>
<tr><td>GSA 18
17.8 M
18.9 F</td></tr>
<tr><td>GSA 19
17.8 M
18.9 F</td></tr>
<tr><td>Aydin & Erdoğan, 2018</td><td colspan="2">Northern Aegean Sea between (Turkey)</td><td>13 F</td><td>April to August</td></tr>
<tr><td>Gherram et al., 2018</td><td colspan="2">Oran Bay (Algeria)</td><td>18.42 M
18.28 F</td><td>January to May</td></tr>
<tr><td>Azzouz et al., 2019</td><td colspan="2">Gulf of Skikda (Algeria)</td><td>14 M
13.65 F</td><td>December-April</td></tr>
<tr><td>Ferreri et al., 2019</td><td colspan="2">Central Mediterranean Sea: Strait of Sicily: Tyrrhenian Sea</td><td>16.1
17.6</td><td>-</td></tr>
<tr><td>Present study</td><td colspan="2">Béni-Saf Bay</td><td>15.6 cm M
14.9 F</td><td>February to July</td></tr>
</table>

the authors attribute this to the temperature of the medium. An increase in the temperatures beyond 11 °C conditions the development of the eggs (Villamor *et al.*, 1997; Wahbi *et al.*, 2015). Our study shows that *Trachurus trachurus* females reached their sexual maturity at 14.9 cm, earlier than males, which attain this maturity at 15.6 cm TL.

To indicate the importance of our results regarding size at first sexual maturity (Lm 50%), the comparative study sets out with different research groups (Tab. 1). Firstly, it is clear that our (Lm 50%) value was much higher than those reported by Aydin & Erdoğan (2018, Northern Aegean Sea near Turkey) and Azzouz *et al*. (2019, Gulf of Skikda, Algeria). On the other hand, the obtained (Lm 50%) value comported fairly well with Viette *et al*. (1997, Italy, Gulf of Trieste), Arneri (1983, Algeria; 1983, Adriatic), and Korichi (1988, Algiers Bay). We found no significant differences compared with research groups of Arruda (1984, Portuguese coast), Abaunza *et al*. (2003, Northwest Atlantic) and Ferreri *et al*. (2019, central Mediterranean Sea), etc. (see Tab. 1).

T. trachurus from Béni-Saf Bay reaches sexual maturity at an earlier size, and this suggests that the fish has changed its sexuality strategy to ensure its sustainability. The species changes the size of its first sexual maturity and reproduces very early.

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OBDOBJE DRSTENJA, SPOLNA ZRELOST IN SPOLNI DELEŽ ŠNJUROV *TRACHURUS TRACHURUS* IZ ZALIVA BÉNI-SAF BAY (ZAHODNA OBALA ALŽIRIJE, JUGOZAHODNO SREDOZEMSKO MORJE)

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POVZETEK

Avtorji so raziskovali razmnoževalne posebnosti pri navadnem šnjuru, Trachurus trachurus *(Linnaeus, 1758), iz zaliva Béni-Saf. V obdobju med novembrom 2015 in oktobrom 2017 so analizirali skupno 355 primerkov, od katerih je bilo 47,04 % samcev, 44,79 % samic in 8,17 % nedoločenih primerkov. Šnjuri so merili med 7,2 in 35,4 cm v dolžino in tehtali od 5,28 do 312,7 g. Spolno zreli samci so merili v dolžino 15,6 cm, spolno zrele samice pa so dosegle 14,9 cm v dolžino. Spremembe v gonadosomatskem indeksu (GSI) so pokazale, da se gonade pri obeh spolih pričnejo razvijati konec februarja, spolno dozorijo pa v maju in juniju, kar opredeljuje obdobje drstenja pri vrsti. Primerki* T. trachurus *iz zaliva Béni-Saf porabljajo prehranske rezerve, pridobljene spomladi, za razvoj spolnih produktov za zgodnje poletni drst.*

Ključne besede: navadni šnjur, *Trachurus trachurus*, razmnoževanje, Béni-Saf Bay, Alžirija

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ON THE OCCURRENCE OF *PSEUDOCARANX DENTEX* (CARANGIDAE) IN THE TURKISH AEGEAN SEA (EASTERN MEDITERRANEAN SEA)

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ABSTRACT

This paper aims to complement and update the data regarding the distribution of uncommon Pseudocaranx dentex *throughout the Mediterranean Sea; specifically by revealing the extension of its distribution in the eastern Mediterranean Sea concerning its habitat preference, such as FADs, sea-cage fish farms, and reefs. A total of 86 specimens of* P. dentex *were observed and one specimen was caught during the period of 2009-2019 from the Aegean Sea. These represent the first well-documented records of* P. dentex *along the south-eastern coast of the Aegean Sea. Their length ranged from 10 to 70.2 cm in TL. Furthermore, the largest* P. dentex *(702 mm TL) found in the Mediterranean so far was recorded during this study.*

Key words: White trevally, FADs, sea-cages, size, habitat

PRESENZA DI *PSEUDOCARANX DENTEX* (CARANGIDAE) NEL MAR EGEO DELLA TURCHIA (MEDITERRANEO ORIENTALE)

SINTESI

L'articolo ha lo scopo di integrare e aggiornare i dati relativi alla distribuzione nel mare Mediterraneo del carango, Pseudocaranx dentex*, specie non comune, rivelando l'estensione della sua distribuzione nel Mediterraneo orientale e considerando le sue preferenze di habitat, come i dispositivi di aggregazione dei pesci (FAD), gli allevamenti ittici in gabbia e le scogliere. In totale sono stati osservati 86 esemplari di* P. dentex*, e un esemplare è stato catturato nel periodo 2009-2019 nel mar Egeo. Questi avvistamenti rappresentano i primi dati ben documentati di* P. dentex *lungo la costa sud-orientale dell'Egeo. La lunghezza degli esemplari variava da 10 a 70,2 cm in lunghezza totale (TL). L'esemplare più grande di* P. dentex *(702 mm TL) trovato finora nel Mediterraneo, è stato avvistato durante questo studio.*

Parole chiave: carango, FAD, gabbie d'allevamento, dimensioni, habitat

INTRODUCTION

White trevally, *Pseudocaranx dentex* (Bloch and Schneider, 1801), is a pelagic and reef-associated species which prefers rocky, hard bottom habitats in tropical (40ºN - 47ºS) seas (Froese & Pauly, 2019). They are generally schooling species but often swim in small groups or solitary at depths from 5 to 238 meters, and they feed on zooplankton and benthic invertebrates (Golani *et al.*, 2006; Bariche, 2012; Froese & Pauly, 2019). Juveniles, in particular, usually inhabit estuaries and shallow waters. Adults form schools and are often associated with rocky bottoms on the continental shelf (Tiralongo, 2018). The common length is 40 cm TL, and the reported maximum length and weight were 122 cm TL and 18.1 kg (Froese & Pauly, 2019).

P. dentex is cosmopolite in tropical and subtropical seas (Golani *et al.*, 2006). In the Mediterranean Sea, *P. dentex* is an uncommon carangid species (Smith-Vaniz, 1986; Bariche, 2012; Tiralongo *et al.*, 2018).

The present paper aims to report on the presence of *P. dentex* throughout the Mediterranean Sea in order to extend the information about its distribution in the eastern Mediterranean Sea concerning its habitat preference, such as Fish Aggregation Devices (FADs), sea-cage fish farms, and reefs. Additionally, the present paper reports the largest *P. dentex* (702 mm TL) recorded to date in the Mediterranean Sea.

MATERIAL AND METHODS

The available information about *P. dentex* was compiled from underwater observations via visual census of wild fish aggregations around experimental FADs and sea-cage fish farms between 2009 and 2017 (Fig. 1A, B and Fig. 2). On August 24th 2019, a specimen of *P. dentex* with a total length (TL) of 702 mm (Fig. 1C) was captured by a spear fisherman in Mersincik Islet, Gökova Bay (Fig. 2) at a depth of 38 m on a rocky bottom (Coordinates: 36°46.046′ N-27°28.353′ E). This specimen was stored in a freezer.



Fig. 1: (A)** Pseudocaranx dentex **around the sea-cages (photo: A. Özgül); (B) around the FADs (photo: A. Özgül); (C) captured from Mersincik islet, Gökova Bay (photo: O. Akyol).

Sl. 1: (A) Trnoboki okoli ribje kletke (foto: A. Özgül); (B) okoli FAD (foto: A. Özgül); (C) ujet primerek iz otočka Mersincik islet, zaliv Gökova (foto: O. Akyol).

RESULTS AND DISCUSSION

A total of 86 specimens (of which 36 from FADs) of *P. dentex* were observed and one specimen was caught in the period of 2009-2019 from the Aegean Sea. These specimens represent the first well-documented records of *P. dentex* in the south-eastern coastal waters of the Aegean Sea. Their length ranged from 10 to 70.2 cm in TL. Diagnostic characters were identified. All details of the specimens are summarized in Table 1. The colour was greenish blue above, silvery white below, with a yellow stripe along the middle of the sides, and a large black spot on the opercula. The morphometric measurements as a percentage of total length (TL %) and the meristic counts recorded in *P. dentex* captured from Gökova Bay, Aegean Sea, are shown in Table 2. All measurements, counts, and colour patterns determined are in accordance with the descriptions of Smith-Vaniz (1986), Golani *et al*. (2006) and Froese & Pauly (2019).

In the Mediterranean Sea, *P. dentex* has been reported on some fish checklists in the Levant Basin (Gücü & Bingel, 1994; Golani, 1996; Saad, 2005; Akel & Karachle, 2017) as well as the Cyclades archipelago (Giokoumi & Kokkoris, 2013) and the coasts of Izmir (as *Caranx dentex*, Geldiay, 1969). Moreover, *P. dentex* has been recorded in the Adriatic Sea since 1986, and one specimen of 227 mm TL was recently caught by a trammel net near Vis Island at a depth of 20 m (Dulcic *et al*., 2003). Also, three fish with lengths ranging from 30 to 40 cm TL have been recorded in the south-eastern coasts of Sicily between Siracusa and Avola (Tiralongo *et al.,* 2018).

P. dentex is considered an uncommon fish, and it might occasionally be caught near to shore with trammel nets or gillnets throughout the Mediterranean. Although *P. dentex* appears to be sporadic in the Adriatic and Levantine Seas, it is relatively more common around the FADs and sea-cage fish farms. Afonso *et al*. (2008) determined that *P. dentex* matures at about 30 cm FL, and its spawning season occurred between June and September in the Azores. The juveniles (12-18 cm) especially preferred the FADs at depths of 50 m, whereas, in the deeper FAD area at depths of 100 m they were not observed. Namely, the juveniles preferred shoreline. On the other hand, those fish from the smallest (10 cm) to the larger (30 cm) aggregated around the sea-cage farms (see Table 1). Here, the contribution of the pellet feed must also be attractive as well as sea-cages that act like mega FADs.

However, the largest specimen with 70.2 cm TL was captured with a spear gun over a rocky bottom. This was the largest size of *P. dentex* that had been observed throughout the Mediterranean. This mature specimen and retinue may have been in the course of reproductive migration: Afonso *et al*. (2008) stated that offshore reefs were a preferential spawning habitat for larger *P. dentex*. At the same time, the capture site was close to a sea-cage fish farm area (i.e., under Güllük Bay), so, the huge specimen had probably reached such a large size due to the high nutritional opportunity around the fish farms.



Fig. 2: The map shows occurring sites of** Pseudocaranx dentex **in the Aegean Sea (□: FADs; ○: sea-cages; red star indicates the sampling location of the huge specimen).
Sl. 2: Zemljevid prikazuje lokalitete, kjer se pojavlja trnobok v Egejskem morju (□: FADs; ○: ribje kletke; rdeča zvezdica označuje vzorčevalno postajo, kjer je bil opažen orjaški primerek).

In fact, *P. dentex* is very rare in the Aegean Sea (only 86 fish during the 17 months in 2009-2019); it is obvious that they are aggregating more where there are existing FADs and/or sea-cage farms. Furthermore, this tropical fish is becoming more abundant due to the effects of global warming in the Mediterranean marine waters (Francour *et al.,* 1994). As indicators of warming in the marine environment, Azzurro (2008)

Tab. 1: Date, location, habitat, depth, distance to land, number of specimens and size range of *Pseudocaranx dentex* **in the Aegean Sea. (*it is caught only one sample).**
Tab. 1: Datumi, lokalitete, habitat, globina, oddaljenost od kopnega, število primerkov in velikostni razpon trnobokov v Egejskem morju (* samo en primerek ulovljen).

Date	Coordinates	Habitat	Depth (m)	Distance to land (m)	Number	TL (cm)	Time of observation
July 2009	38°03´11″N-26°59´01″E	FADs	50	2037	4	14	Daytime
Oct. 2009	38°03´11″N-26°59´01″E	FADs	50	2037	2	12	Daytime
Dec. 2009	38°03´11″N-26°59´01″E	FADs	50	2037	6	12	Daytime
Jan. 2010	38°03´11″N-26°59´01″E	FADs	50	2037	3	12	Daytime
Feb. 2010	38°03´11″N-26°59´01″E	FADs	50	2037	4	14	Daytime
Mar. 2010	38°03´11″N-26°59´01″E	FADs	50	2037	5	12	Daytime
Apr. 2010	38°03´11″N-26°59´01″E	FADs	50	2037	4	15	Daytime
May 2010	38°03´11″N-26°59´01″E	FADs	50	2037	4	15	Daytime
Jun. 2010	38°03´11″N-26°59´01″E	FADs	50	2037	2	16	Daytime
July 2010	38°03´11″N-26°59´01″E	FADs	50	2037	2	18	Daytime
30 June 2016	37°10′49″N-27°22′48″E	Sea-cage	60	1500	27	12-20	08:45
26 June 2016	37°10′49′'N-27°22′48′'E	Sea-cage	60	1500	2	18	09:00
28 Oct. 2016	37°10′49′'N-27°22′48′'E	Sea-cage	60	1500	5	25-30	09:30
23 Dec. 2016	37°10′49′'N-27°22′48′'E	Sea-cage	60	1500	2	30	10:30
12 Apr. 2017	37°10′49′'N-27°22′48′'E	Sea-cage	60	1500	8	10	10:10
15 June 2017	37°17′19′'N-27°24′02′'E	Sea-cage	50	3000	2	25	10:30
24 Aug. 2019	36°46.046'N-27°28.353'E	Rocky	38	550	5*	70.2	Daytime

Tab. 2: Morphometric measurements as percentage of total length (TL %) and meristic counts recorded in the *Pseudocaranx dentex* **captured from Gökova Bay, Aegean Sea.**
Tab. 2: Morfometrične meritve kot odstotek celotne dolžine (TL %) in meristična štetja za trnoboka, ki so ga ujeli v zalivu Gökova v Egejskem morju.

Measurements	Size (mm)	Proportion (TL%)
Total length (TL)	702	
Fork length (FL)	598	85.2
Standard length (SL)	568	80.9
Maximum body depth	195	27.8
Pectoral fin length	185	26.4
Pre-dorsal fin length	230	32.8
Pre-anal fin length	312	44.4
Pre-pectoral length	190	27.1
Head length	181	25.8
Eye diameter	25	3.6
Preorbitary length	79	11.3
Meristic counts		
1st Dorsal fin rays	VIII	
2nd Dorsal fin rays	I+26	
Anal fin rays	II+I+22	
Pectoral fin rays	20	
Ventral fin rays	I+5	
Weight (g)	4126	

concluded that the thermophilic tropical and subtropical fishes such as *Epinephelus marginatus, Caranx crysos, Balistes capriscus, Pseudocaranx dentex, Solea senegalensis, Sphyraena* spp. have extended their distribution margins by crossing their northernmost or southernmost limits in both Mediterranean and extra-Mediterranean areas.

In conclusion, the Mediterranean Sea is currently becoming warmer, in a manner similar to the waters of the rest of the world (Ben Rais Lasram & Mouillot, 2009). Thus, we can expect an increasing of the rate of introduction exotic and thermophilic species to the Mediterranean. However, further studies are required on overlap between exotic/thermophilic and endemic fish fauna and on their competition.

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O POJAVLJANJU TRNOBOKA *PSEUDOCARANX DENTEX* (CARANGIDAE) V TURŠKEM EGEJSKEM MORJU (VZHODNO SREDOZEMSKO MORJE)

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POVZETEK

Avtorji želijo s prispevkom dopolniti in nadgraditi poznavanje razširjenosti trnoboka v Sredozemskem morju, še posebej z vidika razširjanja vrste v vzhodnem Sredozemskem morju in njenih habitatnih preferenc do FAD (naprav za privabljanje rib), ribjih kletk in umetnih podvodnih grebenov. V obdobju 2009-2019 so opazovali skupno 86 primerkov P. dentex v Egejskem morju, en primerek pa so polovili. Ta potrjuje prvi dobro evidentiran primer pojavljanja trnoboka vzdolž jugovzhodne obale Egejskega morja. Primerki so merili od 10 to 70,2 cm telesne dolžine. Poleg tega je največji primerek trnoboka (70,2 cm telesne dolžine) doslej največji zabeležen primerek v Sredozemlju.

Ključne besede: trnobok, FADs, ribje kletke, velikost, habitat

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JADRANSKA MORSKA FLORA

FLORA MARINA ADRIATICA

ADRIATIC MARINE FLORA

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FIRST REPORT OF AN AEGAGROPILOUS FORM OF *RYTIPHLAEA TINCTORIA* FROM THE LAGOON OF STRUNJAN (GULF OF TRIESTE, NORTHERN ADRIATIC)

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ABSTRACT

The occurrence of an aegagropilous form of the red alga Rytiphlaea tinctoria *in the Stjuža marine lagoon of Strunjan (Gulf of Trieste) has been reported for the first time. The distribution, mean diameter, mean density and morphological structure of this population of* R. tinctoria *are here described. During the study, it was observed that the ball-like form of this species differs from the attached form found on open shores, lacking a holdfast, having a radial arrangement of the branches, and a curled distal part of the branches. No reproductive structures were observed in any of the collected samples. The ball-like form of* Rytiphlaea tinctoria *can be considered as an ecotype.*

Key words: *Rytiphlaea tinctoria,* ball-like form, Stjuža lagoon Strunjan, northern Adriatic

PRIMA SEGNALAZIONE DI *RYTIPHLAEA TINCTORIA* IN FORMA EGAGROPILA NELLA LAGUNA DI STRUGNANO (GOLFO DI TRIESTE, ALTO ADRIATICO)

SINTESI

La nota riporta alcune osservazioni su Rytiphlaea tinctoria *in forma egagropila rinvenuta nella Laguna Schiusa di Strugnano (Golfo di Trieste). Vengono descritte la distribuzione, il diametro, la densità e le caratteristiche morfologiche delle forme a palla di questa specie. Lo studio comparativo tra la forma egagropila e quella fissata, non rinvenuta in Laguna, evidenzia delle differenze in quanto priva di strutture d'attacco, ramificazione radiale e ramuli distali più aggrovigliati. Non si sono osservate alcune strutture riproduttive nei campioni raccolti. La forma egagropila presente nella laguna potrebbe essere considerata un ecotipo di* R. tinctoria.

Parole chiave*:* *Rytiphlaea tinctoria,* forma egagropila, laguna Schiusa Strugnano, Alto Adriatico

INTRODUCTION

The red alga *Rytiphlaea tinctoria* (order Ceramiales, family Rhodomelaceae) is a perennial species and can occur in attached or unattached forms, depending on environmental conditions. This alga usually grows attached on rocky substrate, often covered with a thin sandy layer in sheltered and shaded habitats of the upper infralittoral zone (Calvo & Ragonese 1982). It has been reported from Atlantic European coasts, from Great Britain, Spain and Portugal to northern Africa. It is also widespread in Mediterranean coastal areas. In Slovenian coastal waters, the attached form of *R. tinctoria* has been found in Koper Bay (Avčin *et al*. 1974; Vukovič 1982), Strunjan Bay (Avčin *et al*. 1974; Turk & Vukovič, 1994) and Piran Bay (Vukovič 1980; Munda 1993).

The species *R. tinctoria* was described by Clemente (1807) as *Fucus tinctorius*. It is cited in literature for the presence of a water-soluble red pigment in its plastids, called "ficoamatin" by Kützing (1843). Later, Feldman & Tixier (1947) named this pigment "floridorubin". The type locality of this species is Castillo de Santa Catalina and Puerto de Santa Maria and Cádiz, Andalusia, Spain. The name derives from the Latin "*rytis*" = a wrinkle, "*phloios*" = cortex, referring to the transversely furrowed or striate appearance of the surface, and "*tinctorius*" refers to the fact that the alga was used as a source for red dye (Phillips & De Clerck 2005).

Floating balls or unattached mats of *R. tinctoria* have been found in several locations, although reports of the ball-like form of this species are very limited. According to the available literature, this form of *R. tinctoria* has never before been found in the Gulf of Trieste. One of the most studied unattached, ball-like forms of this species has been reported from the Stagnone Lagoon (Sicily, Italy) by Calvo *et al*. (1981), Calvo & Ragonese (1982), Orestano & Calvo (1985), Mercurio *et al.* (2006) and Bellissimo & Orestano (2014).

Unattached algae that grow in a more or less spherical form as free-floating balls are described by the term *aegagropilous*. This term was first used by Linnaeus (1763) as a specific name for a rolling-ball alga from the Baltic: *Conferva aegagropila* L., sin. of *Cladophora aegagropila* (L.) Rabenh., currently regarded as a synonym of *Aegagropila linnaei* Kützing, published by Kützing (1843) (Calvo & Ragonese 1982). Ball-like forms are also formed by several marine species of *Cladophora*, as well as at least 54 other algae, including 25 red, 18 green and 11 brown algae (Norton & Mathieson, 1983).

The present paper reports the occurrence of extensive ball-like aggregates of *Rytiphlaea tinctoria* (Clem.) C.Ag. in the Stjuža marine lagoon of Strunjan (Gulf of Trieste, northern Adriatic). It aims to provide general information on the extensive development of the mobile, free-living rolling balls of this alga observed in the Stjuža marine lagoon of Strunjan in the spring of 2019. Details on the distribution, morphology and morphological measurements of the ball-like form of this alga are reported. Thus, our results contribute to expanding the current knowledge on the unattached, ball-like form of algal populations in this area. The possible factors that led to the formation of the ball-like form of *R. tinctoria* are discussed.

MATERIAL AND METHODS

Study area

The Strunjan Lagoon is a shallow, semi-enclosed oligotrophic brackish coastal lagoon situated in the eastern part of the Strunjan Bay (45° 31′ 30″ North, 13° 36′ 20″ East) (Fig. 1a and 1b), about approx. 10 hectares in surface area and divided into two sub-basins: a smaller discharge lagoon and the larger, main Stjuža Lagoon of the silted former fish-farming pond. Stjuža (from the Italian "chiusa", meaning 'closed') is the only Slovene marine lagoon; it is not completely natural, rather the result of human activities. For about a half century, it has been an abandoned fish farm. After the construction of a dam over 400 years ago, the bay was artificially closed for the purpose of fishing; the newly created lagoon remained connected with the sea only by three tidal channels. The Stjuža Lagoon is characterized by a meadow consisting predominantly of the sea grasses *Cymodocea nodosa* (Ucria) Ascherson and *Zostera noltei* Hornemann on its margins (Vrišer 2002; Šajna & Kaligarič, 2005). Today, the lagoon area is an important part of the Strunjan Stjuža Nature Reserve, within the Natura 2000 network, the primary objective of which is to preserve biodiversity.

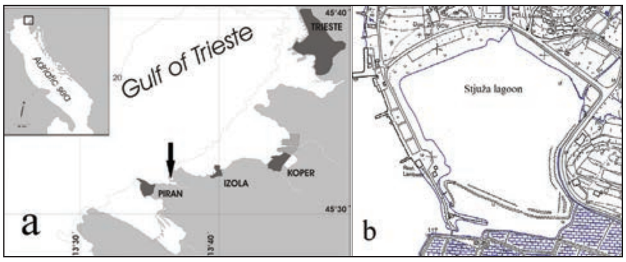


Fig. 1: Maps of the study area (a); Stjuža – Lagoon of Strunjan (b).
Sl. 1: Karta raziskovalnega območja (a); Stjuža – Laguna Strunjan (b).

Environmental parameters

Because of its shallow depth of about 0.5–1 m, the thermal conditions in the Stjuža Lagoon range seasonally from one extreme to the other: between 5 ºC and 10 ºC in wintertime and between 24 ºC and 27 ºC during the summer, while in the other seasons water temperatures are similar to the atmospheric temperatures. Salinity, oxygen content, and thermal conditions in the Stjuža Lagoon

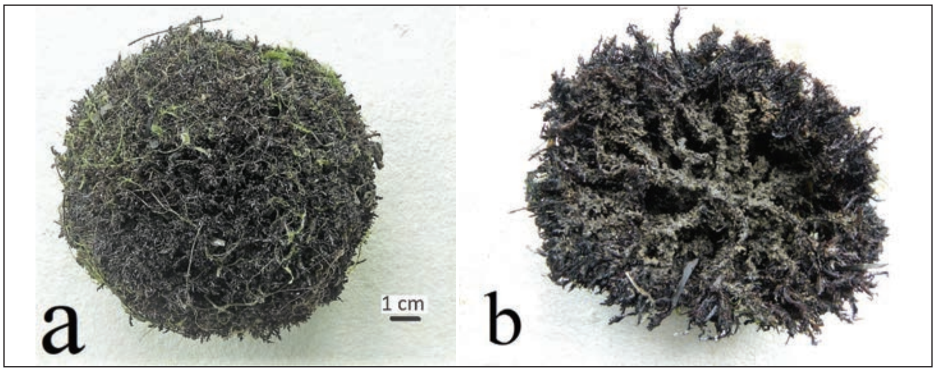


Fig. 2:** Rytiphlaea tinctoria **ball (a); section of** R. tinctoria **ball with branches radially arranged around the centre (b).
Sl. 2: Kroglica alge** Rytiphlaea tinctoria **(a); prerez kroglice alge** R. tinctoria **s prikazom radialno razporejenih poganjkov okoli središča (b).

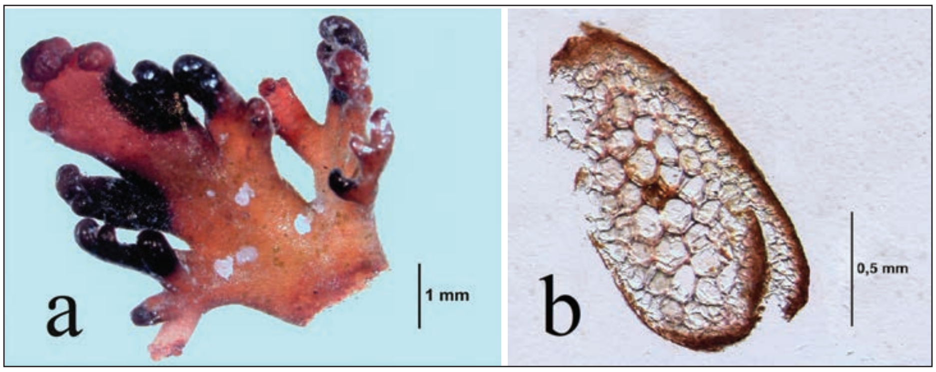


Fig. 3: Branch of the alga** Rytiphlaea tinctoria **with curled apices (a); cross section of a branch showing the axial cell surrounded by 5 pericentral cells with medullary cells and darkly pigmented cortex (b).
Sl. 3: Stranski poganjek alge** Rytiphlaea tinctoria **s kaveljčastimi izrastki (a); prečni prerez poganjka prikaže osrednjo celico, 5 pericentralnih celic, medularne celice in temno pigmentirane kortikalne celice (b).

are related to the large water exchange and are usually similar to those of Strunjan Bay. The lagoon receives freshwater inputs from small canals from agricultural areas (Vrišer, 2002). The average tidal amplitude is 67 cm, with high water reaching 25–45 cm above the mean sea level, and low water 15–30 cm below the mean sea level (Malačič *et al*., 2000).

Sampling procedure and data analysis

The fieldwork was carried out in the spring of 2019, when a dense aggregation of ball-like *R. tinctoria* was found in the Stjuža Lagoon of Strunjan. The study was conducted in separate parts of the lagoon margin on the northern, south-western and western shores. These sites were chosen because of the higher density of ball-like aggregations of this species than in the other parts of the lagoon. The substrate of the entire research area is a soft sediment composed of compact-fine argillaceous silt with a slight admixture of sand, with a thin (0.5–1 cm) yellowish brown layer of flocculent organic detritus (Vrišer, 2002).

The algal material collected was carefully sorted and examined using a stereoscope, while a light microscope was used to check for the presence of reproductive structures. The anatomical observations were based on fresh material. Sections were cut by hand with a single-edged razor blade and photographed in the laboratory of the Natural History Museum of Trieste (Italy) using a microscope Leica MZ16 with camera Leica mc190 HD. All the specimens were identified at a species level, while taxonomically difficult *taxa* were summarized to genus level as 'spp.' due to the absence of reproductive structures and/or diacritical features that are traditionally used to identify a species.

The main resources used to identify the collected species were Maggs & Hommersand (1993), Bressan & Babbini (2003), Phillips & De Clerck (2005), Brodie *et al*. (2007), Sfriso (2010). The nomenclature follows Guiry & Guiry (2019).

Five randomly selected sampling frames (40 cm x 40 cm) were used to estimate the density of the *R. tinctoria* balls. The density was determined by counting the number of the ball-like forms directly from each frame during the field work in each of the three parts of the lagoon checked (northern, south-western and western).

Fifty balls of *R. tinctoria* from the studied area were randomly collected in each of the three parts of the lagoon checked in order to measure their diameter. The measures were taken directly to the nearest 0.1 mm using a caliper.

RESULTS AND DISCUSSION

Dense aggregates of ball-like forms of a red alga were recorded in the Stjuža Lagoon of Strunjan for the first time. Based on morphological features, we identified this red alga as *Rytiphlaea tinctoria*. Its morphology corresponded with the descriptions reported for this species in other parts of the Mediterranean (Phillips & De Clerck, 2005). The balls of *R. tinctoria* ranged in shape from roughly spherical to prolate spheroid (Fig. 2a). Branches were radially arranged around a very small branch segment (Fig. 2b) which formed the core of the thallus.

The distal parts of the thalli consisted of flattened, regularly alternate branches with strongly incurved branch apices. The thallus was yellowish to dark red-brown in colour, darker towards the tips of the axes (Fig. 3a), and coriaceous to cartilaginous in texture, abundantly branched. Microscopic observations showed that the structure of the thallus was uniaxial. Cross section through young and mature branches showed the sub-terete to oval shape of the axes, composed of a central cell surrounded by five pericentral cells, further medullary cells and a darkly pigmented single-layered cortex (Fig. 3b). During our examination of the collected material, we did not detect any reproductive structures.

The values of the mean diameter and mean density of the ball-like *R. tinctoria* from the research area are illustrated in Table 1. The average density was 22.3 (N/1600 cm^2) and it varied between 14 and 33 (N/1600 cm^2). The balls ranged widely in size, their diameter on average 73.5 mm, from a minimum of 36.2 to a maximum of 145.3 mm. The described thalli are morphologically very similar to those described by Calvo & Ragonese (1982) from the Stagnone Lagoon (western coast of Sicily), as shown in Table 1.

The dissection of the balls revealed a solid, dense mass of intertwined branches, almost entirely of *R. tinctoria*, but also containing fragments of degraded material composed mainly of leaves of *C. nodosa* and *Z. noltei*. Some species of Rhodophyta (mainly filamentous Rhodomelaceae with *Polysiphonia*-like morphology), Chlorophyta (mainly Cladophoraceae and *Ulva*) were found as epiphytes on the branches of *R. tinctoria*. In some balls, the only non-organic matter was sand from the substrate. Some balls were kept in an aquarium and numerous small invertebrates were seen emerging from and retreating into the balls, as also reported by Sparla & Riggio (1983-84) and Ballantine *et al*. (1994).

Tab. 1: Average values of the density and comparative size (diameter) recorded for red alga *Rytiphlaea tinctoria* ***balls from the Stjuža Lagoon of Strunjan, and the Stagnone Lagoon (western Sicily).***
Tab. 1: Povprečne vrednosti gostote in primerjalne velikosti (premer) kroglic rdeče alge *Rytiphlaea tinctoria* ***iz Lagune Stjuža Strunjan in Stagnone (zahodna obala Sicilije).***

Rytiphlaea tinctoria			
	Stjuža Lagoon (Strunjan)		**Stagnone Lagoon (Calvo & Ragonese, 1982)**
	Density (N/1600 cm^2)	Size (diameter/ mm)	Size (diameter/mm)
Mean	22.3	73.5	100.0
SD	6.7	24.6	-
Min	14	36.2	40.0
Max	33	145.3	200.0

List of the most abundant epiphytes of *R. tinctoria:*
Ceramium spp.
Chaetomorpha linum (O.F. Müller) Kützing
Cladophora spp.
Cystoseira foeniculacea f. *tenuiramosa (Ercegovic) A. Gómez Garreta, M.C. Barceló, M.A. Ribera & J. Rull Lluch*
Polysiphonia scopulorum Harvey
Polysiphonia spinosa (C. Agardh) J. Agardh
Ulva rigida-laetevirens complex
Titanoderma pustulatum (Lamouroux) Näegeli
Valonia utricularis (Roth) C. Agardh

In the Stjuža Lagoon, many algal species were present in both attached and unattached form. The soft bottom was clearly unsuitable for the development of a highly diverse attached macroalgal vegetation. The presence of the ball-like aggregations of *R. tinctoria* (Fig. 4b) was the consequence of an accumulation of detached material caused by winds and tidal currents flowing during the tidal switch, as illustrated in Fig. 4a, where the yellow arrows indicate the outflow and the red arrows the inflow of the seawater during the change of tides.

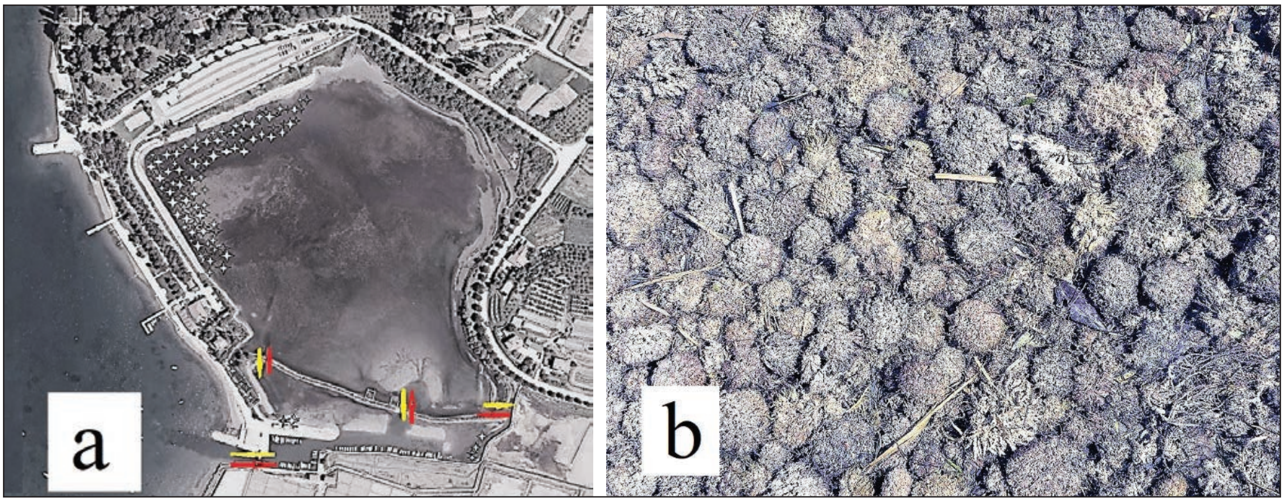


Fig. 4: Distribution of *Rytiphlaea tinctoria* ***ball-form (white stars) occurring in the Stjuža Lagoon of Strunjan and the direction of the currents of the seawater during tidal movement. The yellow arrows indicate the output flow and the red arrows the entry flow (a). Extensive aggregates of*** *Rytiphlaea tinctoria* ***balls (b).***
Sl. 4: Razporeditev kroglic vrste *Rytiphlaea tinctoria* ***(bele zvezdice) v laguni Stjuža v Strunjanu in smer toka morske vode med plimovanjem. Rumene puščice predstavljajo smer izhoda, rdeče pa smer vhoda morske vode med bibavico (a); goste gruče kroglic vrste*** *Rytiphlaea tinctoria* ***(b).***

Due to the lack of hard substrata in the lagoon, we found the attached algae mainly on small pebbles, shells, man-made objects and seagrass rhizomes. Some species were present only in the unattached form, floating above the bottom as benthopleustophytes. We observed only two species of algae with typical ball-like forms: *R. tinctoria* and the green alga *Lychaete echinus* (Biasoletto) M.J. Wynne. Their morphologies were different from those of the attached forms. All unattached macroalgae share some typical morphological features: they lack a basal holdfast and are smaller and more branched than the conspecific attached thalli. They usually exhibit curled or screw-like distal parts of branches.

The pleustophyte populations, rich in ball-like forms (with a free spherical structure) typical of lagoon environments, are frequent in the Mediterranean. Among the ball-like forms typical of lagoon environments in the Mediterranean, *Valonia aegagropila* C. Agardh, *R. tinctoria*, *L. echinus* and *Chaetomorpha linum* (O.F. Müller) Kützing were the most diffused (Calvo *et al*., 1980; Orestano & Calvo, 1985; Cecere *et al*., 1992).

Among the most abundant unattached forms of algae found in the Stjuža Lagoon were green algae of the genus *Ulva*, with the species *U. rigida* and *U. laetevirens* forming mostly unattached accumulations. Sfriso (2006) reported that it would be more correct to refer to the *Ulva rigida-laetevirens* complex, because the two species are indistinguishable when they are in the unattached form. Among these accumulations, *Enteromorpha*-type forms of *Ulva* (with the species *U. compressa* and *U. intestinalis*), *Chaetomorpha* (with the species *C. linum*), *Lychaete* (with the species *L. echinus*) and *Cladophora* (with the species C. *lehmanniana* and *C. liniformis*) were also present.

The brown alga *Cystoseira foeniculacea* f. *tenuiramosa* was detected in two attached forms: as epiphyte on *R. tinctoria* balls (Fig. 5a) and on small pebbles (Fig. 5b). The occurrence of this canopy-forming alga in the Stjuža Lagoon was observed for the first time during this study. Moreover, the species *C. foeniculacea* (Linnaeus) Greville was reported just once in Slovenian coastal waters by Avčin *et al*. (1974) as *Cystoseira discors* (Linnaeus) C. Agardh. Due to the high ecological value of *Cystoseira* spp., this finding is quite important, and the presence and abundance of *C. foeniculacea* f. *tenuiramosa* in the Stjuža Lagoon should be regularly monitored in the future.

Two theories about the formation of the ball-like form of *R. tinctoria* have been proposed. According to the first, the phenomenon can be considered the result of a dynamic action of the waves' motion (Fritsch, 1965; Smith, 1950). The second supports the active role of the alga (Van den Hoek, 1963; Austin, 1960).

Based on the information available for other parts of the Mediterranean, we suggest that some environmental conditions characteristic of the Stjuža Lagoon favour the formation of the ball-like form of *R. tinctoria*, such as: shallowness (with an average of about 0.5–1 m of depth), which permits continuous exposure to sunlight and consequently the growth of algal thalli in all directions; superficial and bottom water currents produced by winds blowing from the North-North-East (burja) and from the South-East (jugo); a wide tidal range, of about 67 cm; and a soft sedimentary bottom unfavourable for the development of attached macroalgae. This is in agreement

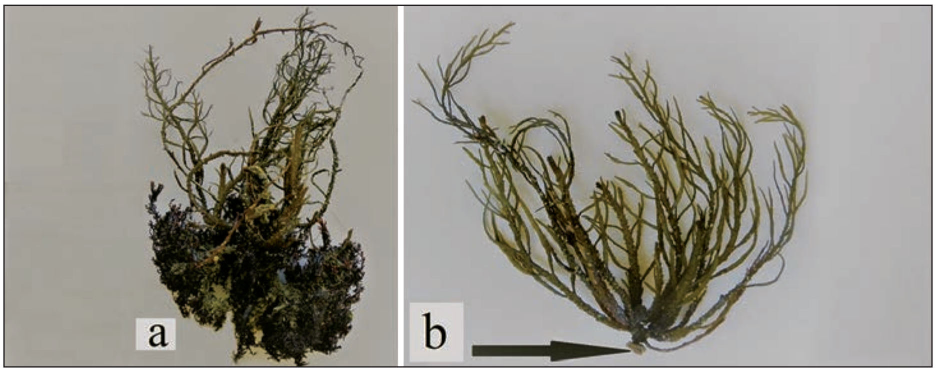


Fig. 5: *Cystoseira foeniculacea* **as epiphyte on** *Rytiphlaea tinctoria* **ball (a); the attached form of** *C. foeniculacea***, the arrow indicates the basal disc (b).**
Sl. 5: *Cystoseira foeniculacea* **kot epifit na kroglici alge** *Rytiphlaea tinctoria* **(a); alga v pritrjeni obliki, puščica prikazuje pritrdilno ploščico.**

with Calvo & Ragonese (1982) and Orestano & Calvo (1985) who, during their studies on the ball-like form of *R. tinctoria* from the Stagnone Lagoon (Sicily, Italy), argued that the formation of the ball-like form is a consequence of two factors: the presence of bottom water currents, which allows rolling, and an intense proliferation of laterals and lateral hooks which, partially imbricate, increase the compactness of the alga. It is generally assumed that the unattached form can develop as spherical, entangled, free-rolling balls under certain hydrographic and topographic conditions. In their study of the formation of ball-like aggregations of the green alga *Aegagropila linnaei* Kützing, Togashi *et al*. (2014) suggested that these aggregations are an adaptative strategy to increase biomass in the extremely limited environments suitable for the growth of this alga.

Another interesting observation made in this study is the absence of reproductive structures in the ball-like form of *R. tinctoria*. It is our general opinion that the benthopleustophyte forms, which derive from the attached form, lost contact with the hard substrate and consequently the capability of development of reproductive cells (Burrows, 1958).

The present observations were limited to a single sampling date. Unfortunately, we did not have more data on the environmental conditions that may have favoured the unusual formation of the ball-like form of this red alga and others, such as *Lychaete echinus* in the Stjuža Lagoon of Strunjan. Our assumptions are based only on the observations made during the short research period and the study of the available literature. It is therefore evident that further investigations, repeated in time, will be necessary for a deeper understanding of this phenomenon.

CONCLUSIONS

On the basis of the cited literature and our observations during the study, we suppose that the formation of the ball-like form of the unattached red alga *R. tinctoria* in the Stjuža Lagoon of Strunjan may be interpreted as a consequence of (a) mechanic processes through a consistent water movement influenced by the winds and tidal current between the high and low tide and (b) features intrinsic to the species which allow the radial growth of the thallus by rolling it on the bottom and thus continuously vary its exposure to light.

On the bases of our field observations on the occurrence of the ball-like form of *R. tinctoria* in the Stjuža Lagoon of Strunjan, which form ball-like aggregations that remain lying or slowly rolling on the bottom, we propose the ball-like form of *R. tinctoria* be considered as an ecotype of this species.

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PRVI ZAPIS O POJAVLJANJU VRSTE *RYTIPHLAEA TINCTORIA* V KROGLIČNI OBLIKI V STRUNJANSKI LAGUNI (TRŽAŠKI ZALIV, SEVERNI JADRAN)

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POVZETEK

Avtorja opisujeta primer prvega pojavljanja kroglične oblike rdeče alge Rytiphlaea tinctoria iz morske lagune Stjuža v Strunjanu (Tržaški zaliv). Terensko delo sta izvedla spomladi 2019 v strunjanski laguni Stjuža, kjer sta opazila visoko gostoto kroglične oblike alge Rytiphlaea tinctoria. Ta se je z večjo gostoto kroglic pojavljala predvsem vzdolž robov lagune na severnem, jugozahodnem in zahodnem delu. Opisujeta porazdelitev, povprečni premer, povprečno gostoto in morfološko zgradbo kroglic R. tinctoria. Za oceno gostote kroglic R. tinctoria sta uporabila pet naključno izbranih kvadrantov (40 cm x 40 cm). Izmerila sta premer petdesetih naključno izbranih kroglic. Ugotovila sta, da je mehko dno lagune za razvoj pritrjene makroalgalne vegetacije očitno neugodno, zato so bile v času študije pritrjene oblike alg prisotne predvsem na majhnih kamenčkih, školjkah, umetnih predmetih in na korenikah morske trave. Avtorja razlagata, da je pojav velike gostote kroglične oblike R. tinctoria v laguni, posledica nanosov, ki so ga povzročili tokovi v laguni zaradi delovanja vetrov in plimovanja. V študiji opisujeta tudi pojav nastanka kroglične oblike R. tinctoria, ki ga skušata razložiti z dvema hipotezama in sicer, pojav lahko obravnavamo kot rezultat dinamičnega delovanja tokov in valov v laguni ali kot rezultat aktivne vloge alge z namenom povečanja biomase v prostorsko omejenem prostoru, kot je laguna. Med študijo sta opazila, da je kroglična oblika te vrste drugačna od značilne pritrjene oblike, ki ni prisotna v laguni, saj nima pritrdilnih struktur in ima radialno razporejene poganjke z ukrivljenimi končnimi deli. V nobenem zbranem vzorcu nista opazila reproduktivnih struktur, kar razlagata kot posledico izgube stika s trdnim substratom in možnostjo za razvoj reproduktivnih struktur. Kroglična oblika Rytiphlaea tinctoria bi lahko veljala za ekotip.

Ključne besede: *Rytiphlaea tinctoria*, kroglična oblika, laguna Stjuža Strunjan, severni Jadran

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SEASONAL GROWTH PATTERNS OF *CYMODOCEA NODOSA* AND DIVERSITY OF ITS EPIBIOTA IN THE NORTHERN ADRIATIC SEA

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ABSTRACT

Growth patterns of the lesser Neptune grass, Cymodocea nodosa*, and the distribution of epifauna and epiflora along its leaves were studied from May to October 2014 at two different depths (1.5 and 5 m) in the northern Adriatic Sea (Pula, Croatia). Generally, seagrass biomass, shoot density and number of leaves per shoot were higher in the shallow water, whereas leaf length was pretty similar between depths. The abundance of epibiota followed a leaf-age gradient. At both depths, autotrophic aufwuchs (mainly Rhodophyta) dominated throughout the study period. The epifauna recorded comprised representatives of Bryozoa, Annelida (Polychaeta), Mollusca and Cnidaria (Anthozoa, Hydrozoa). Above all, we document a major decline of the investigated* C. nodosa *patch from 2014 to the present, which highlights the importance for conservation and management efforts regarding seagrass meadows in the northern Adriatic Sea.*

Key words: aufwuchs, *Cymodocea nodosa,* seagrass meadow, sessile invertebrates

MODELLI DI CRESCITA STAGIONALE DI *CYMODOCEA NODOSA* E DIVERSITÀ DEI SUOI EPIBIONTI NELL'ADRIATICO SETTENTRIONALE

SINTESI

I modelli di crescita di Cymodocea nodosa *e la distribuzione di epifauna ed epiflora lungo le sue foglie sono stati studiati da maggio a ottobre 2014, a due diverse profondità (1,5 e 5 m) nell'Adriatico settentrionale (Pola, Croazia). In generale, la biomassa della pianta, la densità dei fasci e il numero di foglie per fascio sono risultati più alti a 1,5 m, mentre la lunghezza delle foglie era abbastanza simile tra le profondità. L'abbondanza di epibionti ha seguito un gradiente di età delle foglie. Ad entrambe le profondità, "aufwuchs" autotrofi (principalmente Rhodophyta) hanno dominato per tutto il periodo di studio. L'epifauna determinata ha compreso rappresentanti di Bryozoa, Annelida (Polychaeta), Mollusca e Cnidaria (Anthozoa, Hydrozoa). Gli autori hanno inoltre documentato un grave declino del prato di* C. nodosa *nella zona indagata dal 2014 ad oggi, il che evidenzia l'importanza degli sforzi di conservazione e gestione delle praterie di fanerogame nell'Adriatico settentrionale.*

Parole chiave: aufwuchs, *Cymodocea nodosa*, praterie di fanerogame, invertebrati sessili

INTRODUCTION

Seagrass meadows occur across the globe and cover 0.1 - 0.2 % of the oceans (Duarte *et al.*, 2008). Concordant with a general decay of environmental quality in marine near-shore habitats, massive regressions of seagrass meadows have been observed in the last few decades (Zavodnik & Jaklin, 1990; Waycott *et al.*, 2009). This poses a massive threat to coastal ecosystems, as seagrass meadows are important primary producers (Duffy, 2006) and play a crucial role as ecosystem engineers (Wright & Jones, 2006). By colonizing mobile substrata like sandy bottoms or silt at varying depths, they form a three-dimensional structure in an otherwise rather homogeneous environment and supply food, shelter and nursery areas for a variety of animals (Beck *et al.*, 2001; Duffy *et al.,* 2003; Cuadros *et al.*, 2017). Furthermore, seagrasses act as a substrate for a whole community of epiphytes and epizoans (Trautman & Borowitzka, 1999; Piazzi *et al.*, 2016), many of which are strictly associated with the plants and successfully adapted to live and grow on their leaves and rhizomes (Casola *et al.*, 1987; Trautman & Borowitzka, 1999). Despite the general high productivity of seagrass meadows, this aufwuchs plays a crucial role in the seagrass ecosystem functioning by serving as food for a variety of grazing organisms like snails, contributing to the overall productivity and thus representing an important part of the biological diversity in seagrass beds (Silberstein *et al.*, 1986; Moncreiff *et al.*, 1992; Cambridge *et al.*, 2007; Gacia *et al.*, 2009; Lepoint *et al.*, 2014; Piazzi *et al.*, 2016).

Cymodocea nodosa (Ucria) Ascherson, 1870 is one of five seagrass species native to the Adriatic Sea and inhabits mainly sheltered to semi-exposed sites, where it forms either mono-specific or mixed meadows with *Zostera noltei* Hornemann, 1832 (Mazzella *et al.*, 1993; Mazzella *et al.*, 1998). *Cymodocea nodosa* grows at varying salinity conditions down to a depth of 40 m (Procaccini *et al.*, 2003; Boudouresque *et al.*, 2009) and is usually found in places not favoured by the largest growing Mediterranean species, *Posidonia oceanica* (Linnaeus) Delile, 1813 (Toccaceli 1990; Sánchez-Jerez *et al.*, 1999). Considered as a pioneer species, *C. nodosa* is often the first seagrass species to colonize newly established habitat (e.g. Van der Velde & Hartog 1992), but it is also present in degraded *P. oceanica* meadows facilitating the colonisation by other taxa such as the green algae *Caulerpa* spp. (Ceccherelli & Sechi, 2002; Montefalcone *et al.*, 2007). Generally, shoot density and size in seagrasses are mainly determined by meadow depth, health and seasonality, but other factors such as branching frequency, seedlings' success, local sediment type and hydrodynamic conditions are also shaping their growth patterns (Pérez *et al.*, 1994; Marbá *et al.*, 2002; Leoni *et al.*, 2008; Díaz-Almela *et al.*, 2008; Martínez-Crego *et al.*, 2008). Hence, wave action and water currents strongly impact the establishment of *C. nodosa*, whereas in shallow waters (less than 5 m), *C. nodosa* forms patchy meadows that become more continuous in larger depths between 5 and 35 m (Reyes *et al.*, 1995). Maximum levels of biomass and cover are found at intermediate depths (Dring & Dring, 1991; Krause-Jensen *et al.*, 2000).

Despite the important role of *C. nodosa* and its associated community for the functioning of coastal ecosystems (Orlando-Bonaca *et al.*, 2015; 2016), studies about its epifauna and -flora are generally scarce and highly biased towards algal aufwuchs. Previous studies mainly focused on the morphology, diversity, temporal distribution, reproductive phenology, biomass, primary production and distribution of crustose red algae of the family Corallinaceae and other epiphytes on *C. nodosa* leaves (Reyes & Afonso-Carrillo, 1995; Reyes & Sanson, 1996, 1997; Reyes *et al.*, 1998; Reyes & Sansón, 2001). In contrast, next to nothing is known about sessile invertebrates associated with *C. nodosa* (Guidetti *et al.*, 2001). This lack

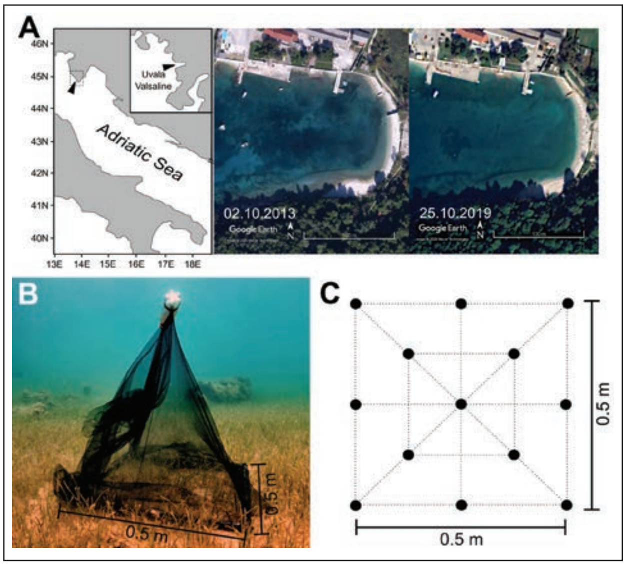


Fig. 1: Sampling. (A) Investigated site Valsaline Bay (Pula, Croatia, 44°50'59.6"N, 13°50'10.0"E). Satellite pictures show a drastic decline in meadow size (dark areas) from 2013 to 2019. (B) Custom-built sampling device with a standardized frame (0.5 m x 0.5 m) attached to the mosquito net. (C) Standardized sampling pattern (black dots) for each replicate (= total of 13 investigated shoots).

Sl. 1: Vzorčenje. (A) Raziskovana lokaliteta – zaliv Valsaline (Pulj, Hrvaška, 44°50'59.6"N, 13°50'10.0"E). Satelitski posnetki kažejo drastični upad v velikosti morskega travnika (temni predeli) od 2013 do 2019. (B) Vzorčevalni pripomoček s standardnim okvirjem (0,5 m x 0,5 m) in mrežico proti komarjem. (C) Standardiziran vzorčevalni pristop (črne točke) za vsako paralelko (=vsota 13 raziskanih šopov).

of data on *C. nodosa* epibiota is quite surprising considering the wealth of studies investigating aufwuchs on *P. oceanica* (e.g. Mazzella *et al.*, 1989; Mazzella *et al*. 1992; Marbá *et al.*, 2002; Balata *et al.*, 2007; Lepoint *et al.*, 2014; Piazzi *et al.*, 2016), which form the basis of our knowledge on seagrass aufwuchs and the factors and processes underlying its diversity and occurrence patterns.

The present study aims at characterizing the seasonal growth patterns of a *C. nodosa* meadow in the northern Adriatic Sea, by comparing two different water depths and relating the growth patterns to different environmental parameters. We further investigated seasonal changes of the diversity and frequency of the whole epibiotic (i.e. autotrophic and heterotrophic) community along the leaves and deliver an overview about the aufwuchs present on *C. nodosa* in the northern Adriatic Sea. Above all, we monitored a major loss of the investigated meadow between the study period (2014) and 2019. The documentation of this decline is of utmost importance for conservation and management efforts regarding *C. nodosa* in the northern Adriatic Sea.

MATERIAL AND METHODS

The investigated *C. nodosa* meadow is located in the northern Adriatic Sea (Valsaline/Pula/Croatia - 44°50′59.6″N 13°50′10.0″E), situated in the southwest of the Istrian peninsula (Fig. 1A). The bay is exposed northwest and some sheltered areas exhibit mixed patches of *C. nodosa* and *Z. noltei*. The present study is focused solely on *C. nodosa* and areas of mixed patches of both species were avoided. Sampling was conducted monthly from May to October 2014 at 1.5 m and 5 m depth by snorkeling and scuba diving. Environmental parameters (surface water temperature, rainfall and wind) were recorded daily during the sampling period and summarized monthly (Tab. 1).

Each monthly sample consisted of four replicates per depth of a standardized area of 0.25 m² where seagrass was collected using a custom-built sampling device (Fig. 1B). For each replicate a smaller frame (14 cm x 14 cm) was used to visually count shoot density and leaves per shoot, which was later extrapolated to a size of 1 m². All other leaves were cut off for calculating the leaf area index (LAI – see below) following Bréda (2008) and kept in seawater with air ventilation for subsequent investigations of epiphytes and epizoans.

$$\frac{\mathrm{T\ DW\ [g]} * \mathrm{RL\ surface\ [mm^2]}}{\mathrm{RL\ DW\ [g]}} = \mathrm{T\ surface\ [mm^2]} \qquad (1)$$

$$\frac{\mathrm{T\ surface\ [m^2]}}{\mathrm{1\ m^2}} = \mathrm{LAI} \qquad (2)$$

where:
T DW = total dry weight [g],
surface T = total surface [mm²], [m²]
RL surface = reference leaf surface [mm²],
RL DW = reference leaf dry weight [g],

In a next step, leaves in a shoot were classified into three categories (*oldest*, *youngest* and *other leaves*). Determination of leaf ages was based on the classification by Reyes & Sansón (2001), with the youngest leaves originating in between the oldest leaves, which typically occupy the outer position in a shoot. Mean leaf lengths were determined for each leaf-age category.

To investigate the epibiota on the leaves, 13 shoots per replicate were harvested in a standardized way (Fig. 1C) (i.e. 52 shoots per month and depth). If possible, aufwuchs was determined to species level. Otherwise higher taxonomical units were used to rank the specimens (Tab. 2). Furthermore, frequency of occurrences as well as total number (i.e. mean and standard deviation) of epibiota on leaves per replicate for taxonomical unit and depth were calculated. For

Tab. 1: Mean and standard deviation (SD) of environmental parameters (water surface temperature, wind speed and direction as well as rainfall) from daily measures during the study period (May-October).
Tab. 1: Povprečje in standardna deviacija (SD) okoljskih parametrov (površinska temperature vode, hitrost in smer vetra ter padavine) na podlagi dnevnih meritev v vzorčevalnem obdobju (maj – oktober).

month	water surface T in °C		wind direction (abundance)								wind speed in km/h			rainfall in l/m²	
	Mean	SD	N	NO	O	SO	S	SW	W	NW	Mean	SD	max	Mean	SD
5	18.07	1.47	9	19	23	18	4	7	10	4	1.36	3.48	75	6.25	3.86
6	21.93	0.98	2	33	19	10	6	6	13	10	11.57	5.12	55	23.00	15.39
7	24.26	0.86	2	35	22	13	2	9	11	11	9.64	2.63	55	15.43	9.32
8	24.90	0.30	7	32	19	20	2	5	9	6	9.21	3.06	50	17.75	5.62
9	21.70	0.65	6	47	21	4	0	4	9	6	1.36	4.02	75	27.17	23.79
10	2.45	0.51	4	37	19	21	3	6	2	4	1.50	5.06	75	9.67	4.51

Tab. 2: Diversity and taxonomic identification of the found epiphytic and epizoic community their abbreviations used in the text and the figures. Insecure taxonomic identifications are marked with "cf." qualifiers.
Tab. 2: Pestrost in taksonomska opredelitev epifitske in epizojske združbe ter njihove okrajšave, uporabljene v slikovnem gradivu in besedilu. Negotove taksonomske določitve so označene s "cf".

Abbreviation	Phylum	Class	Order	Species
cer	Rhodophyta	Florideophyceae	**Ceramiales**	*Ceramium* cf. *diaphanum, Ceramium* cf. *flaccidum, Champia* cf. *parvula, Herposiphonia* cf. *secunda, Laurencia* cf. *minuta, Polysiphonia* cf. *shaerocarpa, Chondria* cf. *mairei*
cor		Florideophyceae	**Corallinales**	*Hydrolithon* cf. *boreale, Hydrolithon* cf. *farinosum, Hydrolithon* cf. *cruciatum, Pneophyllum* cf. *fragile*
rh		diverse **Rhodophyta**		
osc	Cyanobacteria	Cyanophyceae	**Oscillatoriales**	
bac	Ochrophyta	**Bacillariophyceae**	Cocconeidales	
tre	**Foraminifera**	Globothalamea	Rotaliida	*Tretomphaloides concinnus*
for		Tubothalamea	Miliolida	*Massilina* cf. *secans, Peneroplis* cf. *planatus*
schiz	Bryozoa	Gymnolaemata	Cheilostomatida	*Schizobrachiella sanguinea*
bry		Gymnolaemata	Cheilostomatida	*Collarina* cf. *balzaci, Puellina* cf. *gattyae*
pol	Annelida	**Polychaeta**	Sabellida	*Janua* cf. *pagenstecheri, Spirorbis* cf. *borealis, Spirorbis* cf. *corallinae*
qui		**quiver (tube)** of Polychaetes		
biv	Mollusca	**Bivalvia**	Mytiloidea	*Mytilus* cf. *edulis*
ovi		**oviposition** gastropod clutches (possible families: Cerithiidae. Chitonidae. Columbellidae. Conidae. Mangeliidae. Muricidae. Nassariidae. Neridae. Plakobranchidae. Pyramidellidae. Rissoidae. Trochidae)		
buno	Cnidaria	Anthozoa	Actiniaria	***Bunodeopsis strumosa***
cly		Hydrozoa	Leptothecata	***Clytia linearis***
kir		Hydrozoa	Leptothecata	***Kirchenpaueria pinnata***
pach		Hydrozoa	Anthoathecata	***Pachycordyle pusilla***
ATH		Hydrozoa	**Anthoathecata**	

visualizations we decided to show only frequency of occurrences, because they allow a better interspecific comparison between taxa. All graphical visualizations were performed in R v3.3.2 (R Core Team, 2013).

After the investigation period we determined a drastic decline of the seagrass meadow size in Valsaline bay. In order to track this change, we used historical images in GoogleEarthPro. Unfortunately, no high-resolution images were available from 2014. Hence, for visualization purposes (Fig. 1A), we decided to take an aerial image from 2013 and compared it with the situation in 2019. Above all, for a better comparability of the patch size, which can fluctuate seasonally, we show images taken in the same month (October).

RESULTS

Environmental parameters

The annual water surface temperature of the sampling site showed typical seasonal variations in

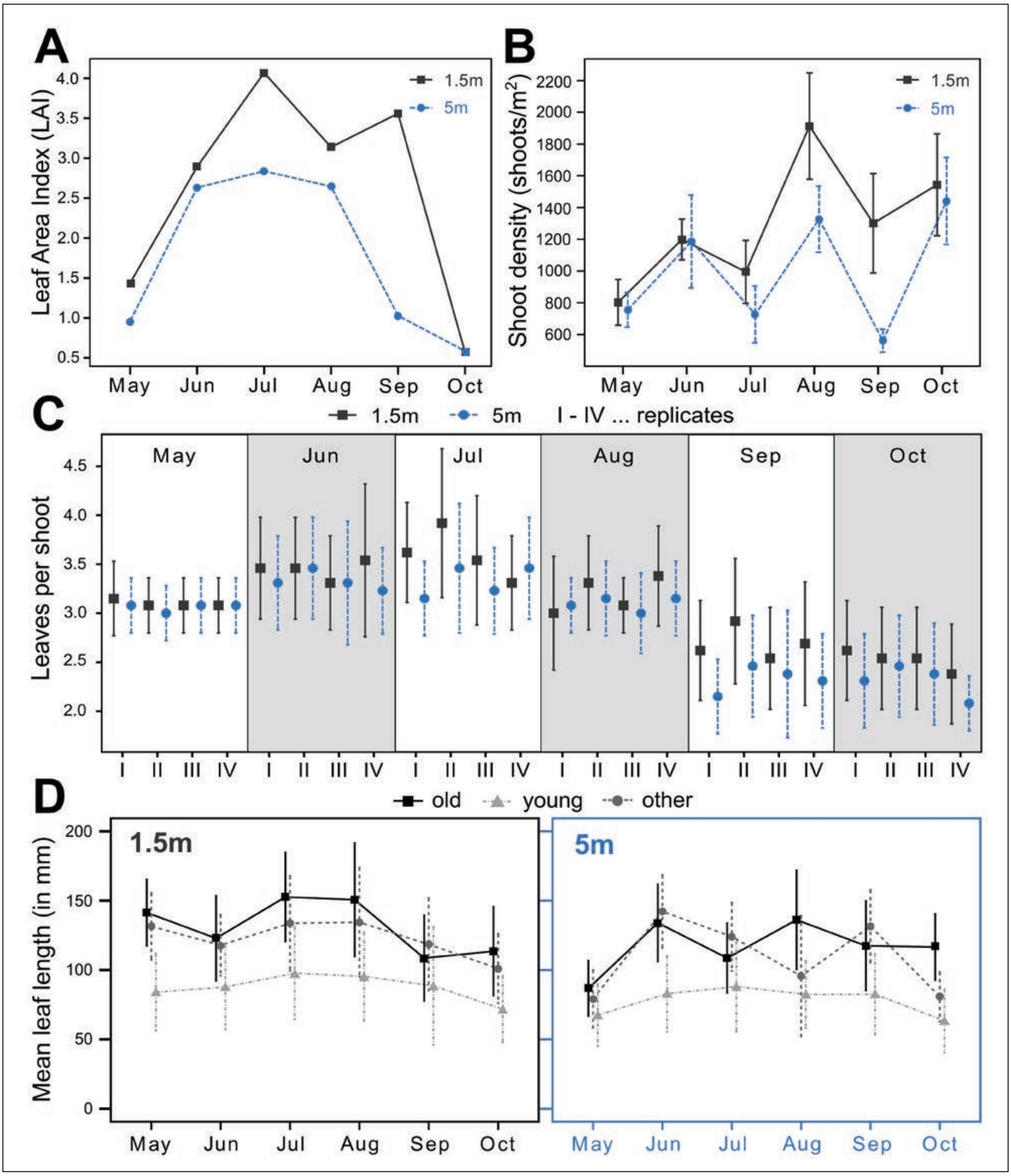


Fig. 2: *Cymodocea nodosa* ***meadow structure of the investigated site (Valsaline, Pula) for the period May - October 2014. (A) Leaf area index (LAI – leaf area/m²), (B) shoot density (shoots/m²), (C) mean number of leaves per shoot for each sampling replicate (I-IV) and (D) comparison of leaf length for different depths, 1.5 m and 5 m.***
Sl. 2: Struktura morskega travnika kolenčaste cimodoceje na raziskani lokaliteti (Valsaline, Pulj) v obdobju med majem in oktobrom 2014. (A) Indeks listne površine (LAI – listna površina/m²), (B) gostota šopov (št. šopov/m²), (C) povprečno število listov v šopu za vsako paralelko (I-IV) in (D) primerjava dolžine listov na različnih globinah 1,5 m in 5 m.

the northern Mediterranean Sea, with a maximum in August (24.9 ± 0.3 °C) and a minimum in May (18.07 ± 1.47 °C) (Tab. 1). The prevailing wind direction was north-east ("Bora"). Average wind speeds did not exceed 11.57 km/h. Maximum wind speed was detected in Mai, September and October with 75 km/h. Highest values of rainfall (27.17 l/m^2) were recorded in September.

Meadow structure

Throughout the study period, seagrass growth parameters were generally higher in 1.5 m compared to 5 m depth (Fig. 2). For both shallow and deep water, the LAI was highest during the summer months, with a maximum LAI observed in July (Fig. 2A). Interestingly, after a drop in August, the LAI in the shallow water reached a second peak in September.

Mean annual shoot density (Fig. 2B) reached values of 1292 ± 433 per m^2 at 1.5 m depth and 1000 ± 385 per m^2 at 5 m depth. Seasonal variation in the number of *C. nodosa* shoots was recorded, with lowest values in May at 1.5 m (803 ± 143 per m^2) and October at 5 m (561 ± 72 per m^2). Highest values were recorded in August at 1.5 m (1913 ± 335 per m^2) and September at 5 m (1441 ± 274 per m^2) (Fig. 2B).

The mean number of leaves per shoot (Fig. 2C) ranged from 3.09 ± 0.41 at 1.5 m to 2.91 ± 0.45 at 5 m. A seasonal pattern became evident, with highest values in July at 1.5 m (3.92 ± 0.76) and in June and July at 5 m (June, 3.46 ± 0.52; July, 3.46 ± 0.66). Lowest values were recorded in October for both depths (1.5 m, 2.38 ± 0.51; 5 m, 2.08 ± 0.28) (Fig. 2C).

The mean leaf length over the entire study period (Fig. 2D) was 131.49 ± 19.15 mm for shallow water and 116.58 ± 18.1 mm for deeper water. The mean leaf lengths (Fig. 2D) of the oldest leaf in a shoot reached maximum values in July (152.61 ± 32.39 mm) and June (133.98 ± 28.23 mm) and minimum values in September (108.56 ± 31.2 mm) and May (86.77 ± 20.36 mm), in 1.5 and 5 m depth, respectively. The maximum values for the youngest leaf in a shoot were recorded in July for both shallow (97.68 ± 33.35 mm) and deeper water (88.03 ± 32.57 mm). Minimum values were observed in October for both depths (1.5 m, 71.78 ± 23.97 mm; 5 m, 63.37 ± 22.65 mm).

Diversity and frequency of epiphytes

A total of 18 taxa were identified, six of which could be determined to species level (Tab. 2). Autotrophic aufwuchs dominated throughout the study period at both depths and was most abundant on the oldest leaves (Fig. 3; Appendix 1, 2 & 3). In contrast, heterotrophic cover did not show

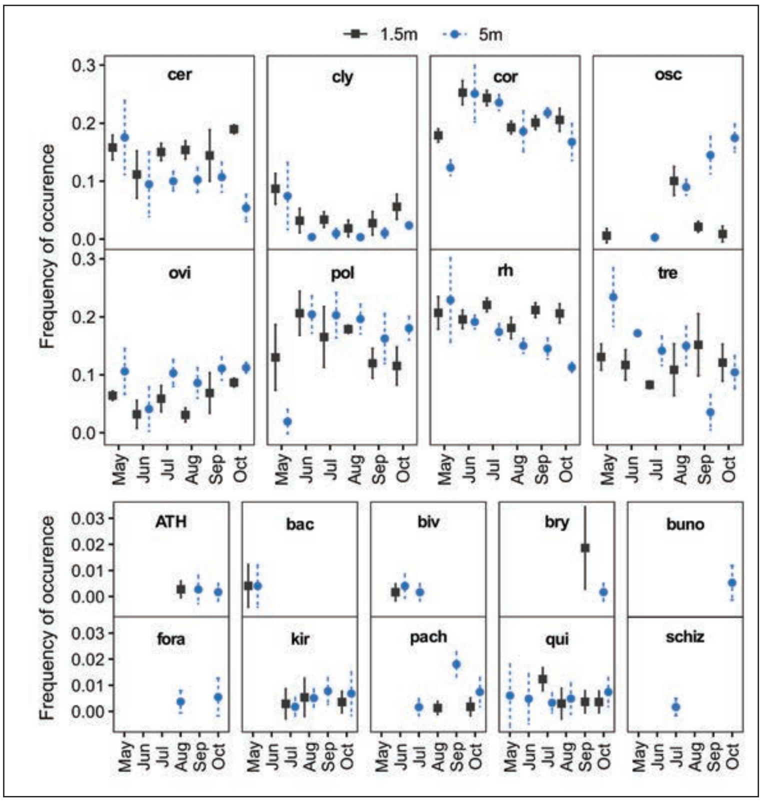


Fig. 3: Frequency of occurrence (mean and standard deviation) of epiphytes and epizoans on leaves per replicate over the investigated period (May – October) for both depths. Abbreviations: cer: *Ceramium*; ***cly:*** *Clytia linearis*; ***cor:*** *Corallinaceae*; ***osc:*** *Oscillatoria*; ***ovi:*** *Oviposition*; ***pol:*** *Polychaeta*; ***rh:*** *Rhodophyta*; ***tre:*** *Tretomphaloides concinna*; ***ATH: a****thecate hydroids*; ***bac:*** *Bacillariophyceae*; ***biv:*** *Bivalvia*; ***bry:*** *Bryozoa*; ***buno:*** *Bunodeopsis strumosa*; ***fora:*** *Foraminifera*; ***kir:*** *Kirchenpaueria pinnata*; ***pach:*** *Pachycordyle pusilla*; ***qui: quiver (tube) of Polychaetes; schiz:*** *Schizobrachiella sanguinea*. ***For taxonomic position see Tab. 2.***

Sl. 3: Frekvenca pojavljanja (povprečje in standardna deviacija) epifitov in epizojev na listih na paralelko v raziskanem obdobju (maj – oktober) na obeh globinah. Okrajšave: cer: *Ceramium*; ***cly:*** *Clytia linearis*; ***cor:*** *Corallinaceae*; ***osc:*** *Oscillatoria*; ***ovi:*** *ovipozicija*; ***pol:*** *Polychaeta*; ***rh:*** *Rhodophyta*; ***tre:*** *Tretomphaloides concinna*; ***ATH:*** *atekatni trdoživnjaki*; ***bac:*** *Bacillariophyceae*; ***biv:*** *Bivalvia*; ***bry:*** *Bryozoa*; ***buno:*** *Bunodeopsis strumosa*; ***fora:*** *Foraminifera*; ***kir:*** *Kirchenpaueria pinnata*; ***pach:*** *Pachycordyle pusilla*; ***qui: cevke mnogoščetincev; schiz:*** *Schizobrachiella sanguinea*. ***Glej Tab. 2. za taksonomski položaj.***

a clear leaf-age gradient (Appendix 2 & 3). In 1.5 m depth, epibiota abundance reached a maximum in July and decreased later in the season (Fig. 3). Similar patterns were observed in 5 m depth, with a maximum in June and July, followed by a decrease later in the study period. *Ceramium* spp., Corallinaceae, various other Rhodophyta, *Tretomphaloides concinna* (Brady, 1884) and Polychaeta were the most frequently observed epibiota at both depth and in all months. Gastropod clutches and *Clytia linearis* (Thorneley, 1900) were present, but at low frequency, in all months at both depth. Oscillatoria, tubes of polychaetes and hydrozoans were present in several months at both depths. Foraminifera, *Schizobrachiella sanguinea* (Norman, 1868), other Bryozoa, Bivalvia, *Bunodeopsis strumosa* Andrès, 1881 were found only occasionally. Most of the hydrozoans, with the exception of *Clytia linearis*, appeared only in the second half (from July onwards) and Bacillariophyceae as well as Bivalvia were only present in the first half (until July) of the season. Taxa that were only present in the deep transect (5 m) included *Bunodeopsis strumosa, Schizobrachiella sanguinea* and various Foraminifera (Fig. 3; Appendix 1).

DISCUSSION

Meadow structure

The structure and overall growth patterns of the studied *C. nodosa* meadow at Valsaline Bay, Croatia, are similar to those of other previously studied *C. nodosa* meadows in the Mediterranean Sea and around the Canary Islands (Bičanić & Baković, 2000; Cancemi *et al.*, 2002; Peduzzi & Vuković, 1990; Reyes *et al.*, 1995). Biomass (LAI) and shoot density were generally higher in 1.5 m as compared to 5 m water depth (Fig. 2A). Previous studies on *C. nodosa* from the Mediterranean Sea and the Canary Islands found maximum growth rates in late spring/summer (Terrados & Ros, 1992; Peduzzi & Vukovic, 1990; Reyes *et al.*, 1995), the time with most favourable conditions for seagrass growth in the Mediterranean Sea (Marbá *et al.*, 1996; Guidetti *et al.*, 2001). Likewise, we found that, at both depths, biomass, shoot density and number of leaves per shoot reached their maxima in the summer months (Fig. 2B & 2C).

Patterns and processes determining meadow structure and seasonal growth patterns of seagrasses are highly complex (Duarte *et al.*, 2007; Garrido *et al.*, 2013), such that disentangling the explicit factors shaping meadow structure and growth patterns is not trivial. *Cymodocea nodosa* is particularly susceptible to seasonal fluctuations (e.g. seawater temperature) and, similar to other seagrasses, the growth pattern is linked to abiotic conditions, particularly to heavy wave action as well as changes in temperature and light intensity (Marbá *et al.*, 1996; Reyes & Afonso-Carrillo, 1995). Mechanic turbulences such as waves could be responsible for discrepancies of growth patterns in different depths. Reduced water movement and an increase of sediment stabilization in deeper water layers enhance seagrass growth and the formation of continuous meadows (Vidondo *et al.*, 1998). In contrast, in shallow water, intense periods of wave action can lead to the fracture of old leaves or detachment of long-living aufwuchs organisms (Reyes & Sansón, 2001). However, we do not find a big difference in overall leaf length when comparing both depths. The investigated site, Valsaline Bay, is opened towards northwest and therefore exposed to western winds (e.g., "Zapadnjak" or "Lebić"). During the study period wind speeds of more than 45 km/h were repeatedly recorded (Tab. 1) from these directions. The prevalent wind direction throughout the study period, however, was north-east ("Bora"). The Bora can reach top speeds of up to 250 km/h (Grisogono *et al.*, 2009) and potentially impacts near-shore seagrass growth patterns. Even though the bay is in general protected from winds of this direction, deeper sites, due to their relative locations in the bay (i.e. larger offshore distance), may be more affected than shallow parts (i.e. closer to shore).

With increasing depth, light availability for photosynthesis decreases (Dring & Dring, 1991; Krause-Jensen *et al.*, 2000), which could explain generally lower LAI and shoot density values (Fig. 2A & 2B) in the deeper water. Furthermore, seagrass meadow structure is known to be influenced by the processes involved in recovering from natural and anthropogenic mechanic stressors (Duarte *et al.*, 2007). Decreasing LAI in August in shallow water could thus be the result of heavy rainfalls in July that discharged large amounts of terrestrial mud into the study area (Tab. 1; S. Bračun, personal observation).

Frequency and diversity of epibiota on *Cymodocea nodosa* leaves

The characteristics of seagrass growth and the life strategies of epiphytes and epizoans are the major factors determining growth dynamics of the aufwuchs (Reyes & Sansón, 1997). A constant creation of leaf surface and the detachment of old parts result in a steadily changing environment that requires adaptations to short life spans and a linear growth/erosion of the substrate (Heijs, 1985; Reyes & Sansón, 2001). In addition, aufwuchs organisms are impacted by numerous abiotic and biotic factors, such as light availability and competition for space

(Heijs, 1985; Reyes & Sansón, 1997), resulting in typical ontogenetic and demographic turnovers. The oldest leaves usually occupy the outermost position of a shoot, and the youngest ones originate from the base between the older leaves of a shoot (Reyes & Sansón, 2001). Initial epibiotic colonizers are found already on the youngest leaves and if they persist and grow, they crucially contribute to an increased biomass of more mature leave stages (Reyes *et al.*, 1998; Reyes & Sansón, 2001).

In our study, autotrophic aufwuchs dominated throughout the study period at both depths (Fig. 3; Appendix 1) and increased with leaf age (Appendix 2 & 3). Reyes & Sansón (2001) showed that the contribution of epiphytes on the oldest leaves, concerning the total biomass of epibiota per shoot, was markedly higher than the contribution of epiphytes from all other leaves. Generally, older leaves are larger (Fig. 2D; Reyes & Sansón, 2001), which offers more leaf surface, but could, especially in nutrient rich areas, affect epiphytic growth due to self-shading effects (Pérez *et al.*, 1994).

Among autotrophic taxa we found various Rhodophyta, Ceramiales (*Ceramium*) and Corallinaceae, all of them quite abundant throughout the year. This is supported by previous studies that recorded Rhodophyta as the most common phylum on seagrass leaves, which account for more than 90% of all algal divisions (Reyes & Sansón, 2001). Among the algal aufwuchs, sciaphilic Corallinaceae were clearly most prominent at both depths (Fig. 3). Crustose coralline algae can bear mechanical disturbances, like strong water movement, are able to grow under low light conditions and are considered as primary colonisers of seagrass leaves (Borowitzka *et al.*, 1990). Whereas in the deep transect (5 m) Corallinaceae could profit from generally lower light conditions, in shallow waters (1.5 m) stronger mechanic disturbances and the shading through leaves resulting from a denser meadow (Fig. 2B) could be explanatory factors for increased Corallinaceae growth.

Overall, epiphyte abundance in shallow water showed an increase until July, followed by a decrease later in the season, and in 5 m depth an increase between June and July, with a subsequent decrease between July and August. Reyes & Sansón (2001) investigated epiphyte biomass on *C. nodosa* leaves throughout the year in the Canary Islands and found an irregular annual variation of the epiphytic community with two maxima (winter, late spring-early summer) and one minimum (spring) that could be linked to the leaf lifetime in different seasons (Reyes *et al.*, 1995). In concordance with these findings, the low frequency of epiphytes in May, detected in our study, might be explained by the short leaf lifetime (45-75 d) (Reyes & Sansón, 1997), which limits the chance of aufwuchs establishment. In contrast, the peak of epiphytes during summer months can be linked to high daily accumulation of epiphytes due to favourable light conditions in this period, and furthermore, could explain the delay of epiphytic growth in deeper water, where these conditions are achieved later in the season. Nonetheless, further studies, especially spanning over winter months, will be necessary to fully understand annual growth dynamics of epiphytes.

Heterotrophic cover appeared seasonally or occasionally, but rarely permanent on *C. nodosa* and did not show a clear leaf-age gradient (Fig. 3; Appendix 2 & 3). Spatial competition could be a reason for the alternating frequency patterns of heterotrophic organisms, with autotrophes potentially suppressing the establishment of sessile epiphytic invertebrates by rapid growth at locations and in months with good light conditions (Borowitzka *et al.*, 1990). Thus, if autotrophic cover decreases, the frequency of heterotrophs can increase. Polychaeta were present throughout the season on leaves of different age, but were most abundant on the youngest leaves, independent of depth (Appendix 2 & 3). Colonisation of young leaves could be a strategy to cope with the enormous space competition in later leaf stages and requires a directed and active settling mechanism. Indeed, sedentary polychaeta larvae have chemotactic organs that might allow them to distinguish between leaf-ages (Pawlik, 1992; Helm *et al.*, 2018). In addition to polychaetes, other organisms such as *Tretomphaloides concinna* (Foraminifera), gastropod clutches and – an exception among Hydrozoans – *Clytia linearis* were present throughout the year. Occasionally found taxa were several Bivalvia, Bacillariophycea, *Bunodeopsis strumosa* and *Schizobrachiella sanguinea*. These occasional occurrences could either be explained by seasonality of organisms (which might be true for athecate hydrozoans, *Kirchenpaueria pinnata* (Linnaeus, 1758) and *Pachycordyle pussila* (Motz-Kossowska, 1905)), or that specimens accidentally settled down on seagrass even though not favorable to them. The sea anemone *Bunodeopsis strumosa* has been reported from different *Zostera* species before (Ates, 1992) and could – as a hemi-sessile animal – easily switch the host plant from adjacent mixed seagrass patches that grow at the investigated study site (Valsaline bay).

Compared to *P. oceanica*, which is considered well-studied regarding its aufwuchs (reviewed by Piazzi *et al.* 2016), the algal communities in *C. nodosa* dominate over sessile invertebrates and appear to be less species-rich (Mazella *et al.*, 1998). Several reasons might explain the discrepancy of auto- versus heterotrophic aufwuchs between *C. nodosa* and *P. oceanica*. Firstly, the size and width of *P. oceanica* leaves largely exceed those of *C. nodosa*, which

allows the establishment of large growing sessile organisms on the leaves. Secondly, *P. oceanica* forms a prominent rhizome layer (also called "matte") that provides settling space and perfect conditions for a variety of sessile invertebrates, many of which can also be found on rocky substrates (e.g. Mabrouk *et al.*, 2014). Hence, these multilayered rhizome structures are unique for *P. oceanica* among all other Mediterranean seagrass species and provide stable perennial conditions that are mostly necessary for invertebrates with longer developmental cycles (Boudouresque, 1974; Piazzi *et al.*, 2016). Several tunicates, poriferan and bryozoan species are associated with the rhizomes of *P. oceanica*, which clearly highlights the pivotal ecological role of this layer for the establishment of heterotrophic aufwuchs (Piazzi *et al.*, 2016). Hence, the absence of a rhizome layer in *C. nodosa* could explain why sponges and tunicates are missing in our samples. However, the whole epibiotic community found in this study is also present in *P. oceanica* (Piazzi *et al.*, 2016). Above all, an overlap in the aufwuchs community between *P. oceanica* and *C. nodosa* makes sense in the light of long-term dynamics and phase-shifts that strongly link these two seagrass species (Montefalcone *et al.*, 2007).

Conservation remarks and outlook

From 2013 to 2019 the investigated seagrass meadow underwent a drastic change, which resulted in the disappearance of the whole patch (Fig. 1A). Several other places along the Istrian coast suffer from a decline of *C. nodosa* meadows (personal observation; L. Lipej, personal communication). Nonetheless, data on distribution and abundance of *C. nodosa* is largely lacking for the northern Adriatic Sea (Orlando-Bonaca *et al.*, 2016), which makes comparative studies and hence the implementation of proper conservation efforts difficult. Natural disturbances (i.e. extreme climatic events, heavy storms, or biological interaction and invasions) are often responsible for seagrass loss, which also affects the biomass and production of its aufwuchs (Reyes & Sansón, 2001; Tuya *et al.*, 2013). On the other hand, human induced disturbances like eutrophication and dredging lead to reduced water clarity and overall quality with impacts on *C. nodosa* growth patterns, or cause direct physical damage (Short & Wyllie-Echeverria, 1996; Reed & Hovel, 2006; Orlando Bonaca *et al.*, 2019). Above all, rapid changes on a global scale affect coastal ecosystems too fast to allow adaptation of seagrass species to the changing environment (Montefalcone *et al.*, 2007; Chefaoui *et al.*, 2018). A loss of seagrass coincides with a deprivation of all the ecological services they provide and as a consequence, water quality, primary production and biodiversity will decline (Tuya *et al.*, 2014).

However, recolonization of seagrass seems still possible if disturbances are limited and growth conditions are suitable. Simulations show that small species of seagrass recover within a few years after a disturbance, while large species may require centuries (Duarte, 1995). Above all, more studies about the dynamics of different seagrass species and their epiphytic community, including *C. nodosa* are essential and of utmost importance for understanding close-shore ecosystem dynamics and changes. Although the knowledge about algal colonization of *C. nodosa* leaves is considered high (e.g. Reyes & Sansón, 1996), there is a lack of data on the diversity of epizoans. Reyes & Sansón (1996) rightly mentioned, that understanding the function of *C. nodosa* comes along with the complete inclusion of the whole aufwuchs community. Hence, further investigations including the creation of a catalogue about the species composition, with proper species determination of all common epifaunal taxa would be necessary, especially in the light of biological diversity and conservation strategies.

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Appendix 1: Frequency of occurrences (upper values) and total number (lower values, grey) of epiphytes on leaves given in mean (bold) and standard deviation (italics) per replicate for both depths. Taxonomic abbreviations correspond to organisms determined in Table 2.

Priloga 1: Frekvenca pojavljanja (zgornje vrednosti) in celotno število (spodnje vrednosti) epifitov in epizojev na listih, izražena s povprečno vrednostjo (krepko) in standardno deviacijo (kurziv) za različne starostne razrede listov na paralelkah v obeh globinah. Taksonomske okrajšave ustrezajo organizmom v Tabeli 2.

	1.5 m						*5 m*					
	May	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**May**	**June**	**Jul**	**Aug**	**Sept**	**Oct**
ATH				**.003** *.003* **2.000** *n.A.*							**.003** *.005* **3.000** *n.A.*	**.002** *.003* **5.000** *n.A.*
bac	**.004** *.008* **1.000** *n.A.*						**.004** *.008* **1.500** *.707*					
biv		**.002** *.003* **1.000** *n.A.*						**.004** *.005* **1.000** *n.A.*	**.002** *.003* **1.000** *n.A.*			
bry					**.019** *.016* **1.200** *.422*							**.002** *.003* **1.000** *n.A.*
buno												**.005** *.007* **1.000** *n.A.*
cer	**.158** *.021* **11.23** *7.681*	**.112** *.041* **1.05** *6.737*	**.150** *.015* **23.91** *1.086*	**.154** *.016* **26.942** *13.616*	**.144** *.044* **4.053** *3.514*	**.190** *.008* **9.714** *8.193*	**.175** *.064* **3.157** *2.619*	**.095** *.056* **3.939** *3.727*	**.100** *.017* **11.951** *8.492*	**.102** *.021* **7.593** *7.025*	**.107** *.026* **2.024** *1.968*	**.054** *.023* **2.500** *2.728*
cly	**.087** *.026* **5.298** *4.573*	**.032** *.021* **3.889** *1.997*	**.034** *.013* **6.318** *3.945*	**.019** *.014* **5.917** *3.895*	**.028** *.020* **8.143** *6.212*	**.056** *.022* **5.290** *3.917*	**.075** *.058* **5.091** *6.131*	**.004** *.004* **5.500** *6.364*	**.010** *.008* **6.667** *12.910*	**.003** *.004* **1.000** *n.A.*	**.010** *.009* **8.000** *6.583*	**.024** *.006* **4.250** *3.545*
cor	**.179** *.011* **19.47** *13.20*	**.253** *.021* **24.99** *13.44*	**.243** *.013* **32.62** *17.38*	**.192** *.011* **34.07** *18.00*	**.201** *.012* **18.33** *9.548*	**.206** *.020* **2.31** *1.78*	**.123** *.013* **3.118** *3.122*	**.251** *.049* **15.60** *1.08*	**.236** *.014* **24.92** *9.329*	**.186** *.035* **22.47** *13.63*	**.218** *.008* **22.512** *9.470*	**.168** *.032* **21.630** *1.447*
fora										**.004** *.004* **1.000** *n.A.*		**.005** *.007* **1.000** *n.A.*
kir			**.003** *.006* **3.500** *.707*	**.005** *.007* **3.000** *3.367*		**.004** *.004* **3.500** *2.121*			**.002** *.004* **2.000** *n.A.*	**.005** *.003* **1.000**	**.008** *.005* **3.000** *2.646*	**.007** *.008* **2.333** *2.309*
osc	**.006** *.012* **4.333** *5.774*			**.100** *.025* **5.044** *4.621*	**.021** *.009* **1.727** *1.555*	**.009** *.014* **1.800** *1.789*			**.003** *.006* **1.000** *n.A.*	**.090** *.013* **5.098** *3.562*	**.145** *.032* **4.196** *2.895*	**.174** *.024* **7.659** *5.114*
ovi	**.064** *.008* **1.486** *1.269*	**.032** *.024* **1.294** *.588*	**.059** *.022* **1.216** *.584*	**.031** *.012* **1.286** *.956*	**.069** *.035* **1.676** *.944*	**.087** *.008* **1.500** *.772*	**.106** *.040* **1.578** *1.011*	**.041** *.038* **1.421** *.838*	**.103** *.023* **1.714** *1.300*	**.086** *.026* **1.780** *1.542*	**.111** *.020* **1.465** *.797*	**.112** *.009* **1.564** *1.014*
pach				**.001** *.003* **1.00** *n.A.*		**.002** *.003* **3.000** *n.A.*			**.002** *.003* **43.00** *n.A.*		**.018** *.005* **15.714** *12.473*	**.007** *.006* **8.750** *6.292*
pol	**.130** *.057* **1.986** *1.467*	**.206** *.038* **3.549** *3.038*	**.165** *.052* **2.819** *2.129*	**.179** *.007* **2.975** *2.270*	**.120** *.026* **2.397** *1.632*	**.115** *.033* **1.641** *1.132*	**.019** *.021* **1.111** *.333*	**.204** *.032* **2.392** *1.781*	**.203** *.039* **3.098** *2.520*	**.196** *.026* **2.634** *1.745*	**.163** *.043* **3.460** *2.669*	**.180** *.021* **3.614** *2.516*
qui			**.012** *.004* **1.000** *n.A.*	**.003** *.006* **1.000** *n.A.*	**.004** *.004* **1.000** *n.A.*	**.004** *.004* **1.000** *n.A.*	**.006** *.012* **1.333** *.577*	**.005** *.010* **1.000** *n.A.*	**.003** *.004* **1.000** *n.A.*	**.005** *.006* **1.000** *n.A.*		**.007** *.006* **5.750** *2.754*
rh	**.207** *.028* **17.55** *12.303*	**.196** *.016* **2.70** *11.99*	**.221** *.012* **29.67** *15.20*	**.181** *.018* **3.39** *15.48*	**.212** *.013* **14.153** *9.721*	**.206** *.016* **8.947** *7.095*	**.229** *.072* **4.242** *2.689*	**.191** *.012* **1.05** *7.100*	**.174** *.014* **17.61** *8.785*	**.150** *.013* **11.33** *9.168*	**.145** *.018* **2.446** *1.972*	**.113** *.008* **2.071** *1.333*
schiz									**.002** *.003* **1.000** *n.A.*			
tre	**.131** *.022* **1.806** *1.241*	**.117** *.026* **2.047** *1.408*	**.083** *.006* **2.019** *1.513*	**.109** *.045* **2.427** *1.839*	**.152** *.053* **2.914** *2.128*	**.121** *.032* **1.597** *.954*	**.234** *.051* **3.340** *3.827*	**.172** *.005* **2.165** *1.396*	**.142** *.025* **1.884** *1.545*	**.150** *.034* **2.058** *1.521*	**.036** *.030* **1.286** *.469*	**.104** *.028* **1.340** *.586*

Appendix 2: Frequency of occurrences (upper values, white) and total number (lower values, grey) of epiphytes on leaves given in mean (bold) and standard deviation (italics) for different leaf-ages per replicate for 1.5 m depth. Taxonomic abbreviations correspond to organisms determined in Table 2.
Priloga 2: Frekvenca pojavljanja (zgornje vrednosti na belem polju) in celotno število (spodnje vrednosti na sivem polju) epifitov in epizojev na listih, izraženi s povprečno vrednostjo (krepko) in standardno deviacijo (kurziv) za različne starostne razrede listov na paralelka v globini 1,5 m. Taksonomske okrajšave ustrezajo organizmom v Tabeli 2.

depth	*1.5 m*																	
leaf-age	***Old***						***Young***						***Others***					
month	May	Jun	Jul	Aug	Sep	Oct	May	Jun	Jul	Aug	Sep	Oct	May	Jun	Jul	Aug	Sep	Oct
ATH				**.003** *.01*												**.004** *.01*		
				2.00 *n.A.*												**2.00** *n.A.*		
bac	**.01** *.02*																	
	1.00 *n.A.*																	
biv		**.004** *.01*																
		1.00 *n.A.*																
bry					**.004** *.01*						**.03** *.04*						**.03** *.03*	
					1.00 *n.A.*						**1.25** *.50*						**1.20** *.45*	
cer	**.22** *.01*	**.17** *.06*	**.22** *.02*	**.18** *.02*	**.16** *.06*	**.20** *.01*	**.05** *.06*	**.03** *.05*		**.04** *.03*	**.08** *.05*	**.17** *.02*	**.13** *.05*	**.09** *.04*	**.14** *.02*	**.16** *.02*	**.16** *.03*	**.19** *.01*
	13.42 *8.16*	**1.92** *6.88*	**27.40** *1.18*	**3.44** *14.34*	**4.73** *3.98*	**12.52** *8.80*	**11.80** *8.70*	**6.33** *4.93*		**1.25** *5.97*	**2.88** *2.36*	**4.28** *4.59*	**7.21** *4.62*	**9.05** *6.64*	**2.11** *8.58*	**24.49** *11.68*	**3.35** *2.81*	**1.38** *7.29*
cly	**.04** *.01*	**.03** *.02*	**.03** *.02*	**.03** *.02*	**.02** *.02*	**.06** *.03*	**.13** *.08*	**.02** *.03*	**.02** *.03*	**.03** *.04*	**.04** *.01*	**.05** *.04*	**.12** *.05*	**.04** *.03*	**.04** *.02*	**.01** *.01*	**.03** *.04*	**.06** *.04*
	2.80 *2.94*	**3.29** *1.50*	**8.43** *4.12*	**5.43** *3.46*	**7.00** *6.87*	**4.79** *3.40*	**5.25** *5.53*	**2.00** *1.41*	**4.50** *.71*	**8.33** *5.51*	**7.25** *6.70*	**6.00** *5.32*	**6.32** *4.38*	**4.78** *2.11*	**5.46** *3.84*	**4.00** *2.83*	**1.75** *5.56*	**5.44** *3.61*
cor	**.22** *.01*	**.24** *.03*	**.23** *.01*	**.18** *.02*	**.20** *.01*	**.21** *.02*	**.07** *.06*	**.18** *.05*	**.27** *.07*	**.21** *.05*	**.17** *.07*	**.21** *.04*	**.18** *.03*	**.29** *.03*	**.25** *.01*	**.20** *.01*	**.22** *.02*	**.20** *.01*
	28.06 *11.54*	**35.73** *11.37*	**46.75** *15.12*	**47.17** *15.09*	**21.98** *7.06*	**28.04** *8.65*	**9.14** *3.80*	**11.38** *7.89*	**15.46** *7.28*	**12.70** *5.58*	**9.00** *8.88*	**9.56** *4.82*	**9.87** *6.97*	**2.27** *9.89*	**28.53** *13.80*	**29.61** *13.42*	**17.97** *9.78*	**2.04** *7.23*
kir				**.01** *.01*					**.01** *.02*			**.01** *.01*			**.003** *.01*	**.01** *.01*		**.01** *.01*
				5.00 *4.24*					**4.00** *n.A.*			**5.00** *n.A.*			**3.00** *n.A.*	**1.00** *n.A.*		**2.00** *n.A.*
osc	**.01** *.02*			**.12** *.03*	**.04** *.02*	**.01** *.02*				**.02** *.02*	**.01** *.02*	**.01** *.02*	**.01** *.01*			**.11** *.04*		
	6.00 *7.07*			**4.43** *3.74*	**1.80** *1.62*	**1.00** *n.A.*				**2.50** *.71*	**1.00** *n.A.*	**3.00** *2.83*	**1.00** *n.A.*			**5.90** *5.50*		
ovi	**.06** *.02*	**.02** *.03*	**.05** *.02*	**.05** *.02*	**.07** *.04*	**.07** *.04*	**.06** *.06*	**.01** *.02*	**.08** *.05*	**.03** *.04*	**.05** *.04*	**.08** *.03*	**.08** *.03*	**.04** *.03*	**.06** *.03*	**.02** *.01*	**.09** *.03*	**.13** *.04*
	1.29 *.61*	**1.20** *.45*	**1.08** *.29*	**1.31** *1.11*	**1.65** *.70*	**1.53** *.80*	**1.20** *.45*	**1.00** *n.A.*	**1.43** *.53*	**1.00** *n.A.*	**1.80** *1.79*	**1.36** *.63*	**1.75** *1.77*	**1.36** *.67*	**1.22** *.73*	**1.40** *.89*	**1.67** *.90*	**1.59** *.87*
pach						**.004** *.01*										**.003** *.01*		
						3.00 *n.A.*										**1.00** *n.A.*		
pol	**.08** *.04*	**.15** *.04*	**.12** *.04*	**.14** *.02*	**.14** *.04*	**.11** *.02*	**.24** *.11*	**.28** *.07*	**.22** *.12*	**.27** *.06*	**.06** *.07*	**.12** *.05*	**.14** *.06*	**.23** *.04*	**.18** *.05*	**.19** *.02*	**.13** *.04*	**.11** *.04*
	1.44 *.98*	**3.28** *3.05*	**2.70** *1.92*	**2.79** *2.03*	**2.51** *1.72*	**1.54** *.88*	**1.77** *1.11*	**2.58** *2.30*	**2.14** *1.59*	**1.96** *1.45*	**1.71** *1.11*	**1.86** *1.62*	**2.42** *1.77*	**4.11** *3.22*	**3.12** *2.35*	**3.61** *2.57*	**2.43** *1.63*	**1.53** *.64*
qui			**.02** *.02*	**.01** *.01*	**.01** *.01*	**.004** *.01*						**.01** *.01*			**.01** *.01*			
			1.00 *n.A.*	**1.00** *n.A.*	**1.00** *n.A.*	**1.00** *n.A.*						**1.00** *n.A.*			**1.00** *n.A.*			
rh	**.22** *.01*	**.24** *.03*	**.23** *.01*	**.18** *.02*	**.20** *.01*	**.21** *.02*	**.15** *.08*	**.11** *.09*	**.19** *.05*	**.13** *.07*	**.22** *.07*	**.21** *.04*	**.22** *.05*	**.19** *.04*	**.23** *.01*	**.19** *.01*	**.22** *.02*	**.20** *.01*
	26.56 *11.19*	**24.59** *11.17*	**39.52** *13.88*	**38.69** *13.93*	**17.12** *8.72*	**1.96** *7.64*	**6.20** *4.00*	**12.82** *7.25*	**14.12** *5.77*	**11.77** *5.56*	**7.21** *8.13*	**5.22** *4.70*	**11.07** *7.08*	**18.22** *12.39*	**26.24** *13.00*	**27.00** *13.32*	**14.58** *9.95*	**1.11** *6.94*
tre	**.15** *.04*	**.15** *.02*	**.11** *.03*	**.10** *.04*	**.16** *.04*	**.13** *.02*	**.09** *.05*	**.04** *.03*	**.04** *.03*	**.11** *.12*	**.18** *.14*	**.13** *.04*	**.12** *.01*	**.12** *.03*	**.07** *.02*	**.11** *.05*	**.12** *.03*	**.09** *.05*
	1.95 *1.31*	**2.59** *1.62*	**2.19** *1.44*	**2.07** *1.27*	**3.24** *2.33*	**1.79** *1.08*	**1.22** *.44*	**1.75** *.96*	**1.00** *n.A.*	**3.31** *2.84*	**2.10** *1.70*	**1.18** *.39*	**1.81** *1.30*	**1.46** *.88*	**2.00** *1.68*	**2.38** *1.71*	**3.11** *1.94*	**1.83** *1.11*

Appendix 3: Frequency of occurrences (upper values) and total number (lower values) of epiphytes on leaves given in mean (bold) and standard deviation (italics) for different leaf-ages per replicate for 5 m depth. Taxonomic abbreviations correspond to organisms determined in Table 2.
Priloga 3: Frekvenca pojavljanja (zgornje vrednosti) in celotno število (spodnje vrednosti) epifitov in epizojev na listih, izraženi s povprečno vrednostjo (krepko) in standardno deviacijo (kurziv) za različne starostne razrede listov na paralelka v globini 5 m. Taksonomske okrajšave ustrezajo organizmom v Tabeli 2.

depth	5 m																	
leaf-age	*Old*						*Young*						*Others*					
month	May	Jun	Jul	Aug	Sep	Oct	May	Jun	Jul	Aug	Sep	Oct	May	Jun	Jul	Aug	Sep	Oct
ATH					**.01** *.01* **3.00** *n.A.*													**.01** *.02* **5.00** *n.A.*
bac	**.01** *.02* **1.50** *.71*																	
biv		**.004** *.01* **1.00** *n.A.*												**.005** *.01*	**.004** *.01*			
bry						**.003** *.01*												
buno						**.01** *.01*												
cer	**.21** *.05* **4.08** *3.10*	**.15** *.07* **4.36** *4.11*	**.14** *.02* **12.9** *8.80*	**.14** *.02* **7.67** *6.85*	**.10** *.02* **2.32** *1.89*	**.05** *.03* **3.06** *3.23*	**.09** *.10* **2.14** *1.21*	**.03** *.03* **1.00** *n.A.*	**.02** *.03* **2.00** *1.41*	**.01** *.02* **1.00** *n.A.*	**.10** *.06* **1.27** *.47*	**.06** *.02* **1.88** *2.10*	**.18** *.08* **2.04** *1.27*	**.06** *.06* **3.36** *2.76*	**.09** *.02* **11.3** *7.94*	**.09** *.03* **7.81** *7.73*	**.16** *.10* **2.25** *3.15*	**.05** *.04* **1.50** *.58*
cly	**.04** *.04* **7.13** *6.75*		**.02** *.01* **9.25** *15.8*	**.01** *.01* **1.00** *n.A.*	**.01** *.01* **2.50** *2.12*	**.03** *.01* **5.57** *4.08*	**.14** *.18* **3.36** *2.58*						**.09** *.06* **5.29** *7.60*	**.01** *.01* **5.50** *6.36*	**.01** *.01* **1.50** *.71*		**.03** *.04* **13.5** *2.12*	**.05** *.06* **2.40** *1.52*
cor	**.16** *.04* **3.39** *2.68*	**.25** *.05* **2.8** *8.98*	**.21** *.03* **3.0** *7.17*	**.17** *.02* **32.6** *9.50*	**.25** *.03* **25.4** *8.30*	**.18** *.03* **27.6** *7.83*	**.08** *.03* **2.00** *1.00*	**.15** *.07* **4.30** *4.30*	**.26** *.08* **14.4** *6.47*	**.16** *.07* **1.9** *4.02*	**.16** *.03* **15.4** *5.85*	**.12** *.04* **1.2** *6.17*	**.09** *.02* **3.08** *4.61*	**.28** *.06* **12.9** *9.10*	**.25** *.02* **24.9** *8.43*	**.22** *.07* **13.6** *1.6*	**.20** *.05* **21.2** *12.5*	**.21** *.05* **14.2** *5.34*
fora				**.01** *.01* **1.00** *n.A.*		**.01** *.01* **1.00** *n.A.*						**.01** *.01* **1.00** *n.A.*						
kir				**.01** *.01* **1.00** *n.A.*	**.004** *.01* **1.00** *n.A.*	**.01** *.01* **3.00** *2.83*					**.01** *.02* **2.00** *n.A.*				**.004** *.01* **2.00** *n.A.*		**.01** *.02* **6.00** *n.A.*	**.01** *.03* **1.00** *n.A.*
osc			**.004** *.01* **1.00** *n.A.*	**.13** *.01* **5.00** *3.62*	**.18** *.03* **4.63** *2.84*	**.18** *.03* **9.48** *5.35*					**.09** *.07* **2.10** *1.85*	**.17** *.04* **4.39** *2.37*			**.004** *.01* **1.00** *n.A.*	**.07** *.03* **5.38** *3.52*	**.13** *.05* **4.75** *3.33*	**.18** *.05* **6.33** *4.66*
ovi	**.11** *.03* **1.48** *.75*	**.05** *.06* **1.11** *.33*	**.10** *.03* **1.73** *.78*	**.09** *.03* **2.27** *1.93*	**.11** *.04* **1.54** *.83*	**.13** *.01* **1.53** *1.03*	**.09** *.07* **1.13** *.35*	**.03** *.06* **2.50** *.71*	**.11** *.02* **1.30** *.67*	**.09** *.04* **1.00** *n.A.*	**.11** *.03* **1.23** *.44*	**.07** *.03* **1.80** *1.14*	**.11** *.06* **1.94** *1.39*	**.04** *.03* **1.50** *1.07*	**.10** *.03* **1.85** *1.79*	**.08** *.03* **1.40** *.83*	**.11** *.04* **1.67** *1.21*	**.13** *.08* **1.43** *.79*
pach					**.01** *.01* **16.3** *15.0*						**.04** *.01* **15.3** *12.7*	**.01** *.01* **18.0** *n.A.*			**.003** *.01* **43.0** *n.A.*			**.03** *.03* **5.67** *1.53*
pol	**.01** *.01* **1.00** *n.A.*	**.11** *.06* **1.79** *1.28*	**.16** *.02* **3.48** *3.04*	**.12** *.02* **1.97** *1.19*	**.15** *.04* **3.31** *2.92*	**.18** *.02* **3.94** *2.32*	**.04** *.08* **1.00**	**.38** *.13* **2.80** *2.10*	**.28** *.11* **1.65** *.75*	**.38** *.06* **2.94** *1.76*	**.22** *.05* **2.96** *1.66*	**.20** *.03* **2.89** *2.89*	**.03** *.04* **1.20** *.45*	**.24** *.02* **2.47** *1.77*	**.21** *.05* **3.49** *2.44*	**.22** *.07* **2.98** *2.01*	**.09** *.04* **7.00** *3.08*	**.17** *.07* **3.91** *2.26*
qui	**.004** *.01* **1.00** *n.A.*	**.01** *.03* **1.00** *n.A.*	**.004** *.01* **1.00** *n.A.*	**.01** *.01* **1.00** *n.A.*		**.01** *.01* **4.67** *2.08*						**.01** *.01* **9.00** *n.A.*	**.01** *.02* **1.50** *.71*		**.003** *.01*			
rh	**.27** *.08* **5.32** *3.04*	**.24** *.02* **11.2** *7.66*	**.20** *.01* **18.7** *9.41*	**.17** *.02* **13.57** *8.52*	**.13** *.02* **2.52** *1.22*	**.12** *.01* **2.12** *1.45*	**.19** *.14* **3.00** *1.46*	**.07** *.07* **8.80** *3.03*	**.05** *.04* **13.4** *9.24*	**.07** *.06* **3.29** *2.56*	**.12** *.02* **2.36** *1.55*	**.13** *.03* **1.89** *1.18*	**.20** *.06* **3.14** *1.72*	**.18** *.03* **8.80** *6.58*	**.19** *.02* **17.0** *8.09*	**.15** *.03* **9.25** *9.89*	**.23** *.06* **2.40** *3.20*	**.06** *.07* **2.40** *1.14*
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tre	**.19** *.05* *3.46*	**.18** *.02* **2.39** *1.59*	**.15** *.01* **2.08** *1.83*	**.14** *.03* **2.34** *1.91*	**.04** *.04* **1.33** *.50*	**.09** *.05* **1.29** *.53*	**.23** *.10* **2.45** *1.99*	**.10** *.09* **1.29** *.49*	**.14** *.08* **1.15** *.38*	**.20** *.09* **1.84** *1.01*	**.03** *.02* **1.00** *n.A.*	**.12** *.02* **1.35** *.61*	**.29** *.08* **3.23** *4.73*	**.18** *.03* **2.10** *1.26*	**.14** *.03* **1.94** *1.43*	**.14** *.03* **1.77** *1.03*	**.02** *.03* **2.00** *n.A.*	**.09** *.08* **1.50** *.76*

SEZONSKA RAST KOLENČASTE CIMODOCEJE (*CYMODOCEA NODOSA*) IN PESTROST NJENIH EPIBIONTOV V SEVERNEM JADRANU

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POVZETEK

Avtorji so raziskovali rastne značilnosti kolenčaste cimodoceje (Cymodocea nodosa) *in prostorsko poraščenost epifavne in epiflore na njenih listih od maja do oktobra 2014 na dveh globinah (1,5 in 5 m) v severnem Jadranu (Pulj, Hrvaška). Navadno so biomasa, gostota šopov in število listov v šopu višji v plitvini, medtem ko je dolžina lista bolj ali manj podobna na različnih globinah. Število epibiontov je sledilo gradientu starosti listov. Na obeh globinah so tekom raziskave kot obrast na listih prevladovale rdeče alge. Epifavno so sestavljali predstavniki taksonomskih skupin kot so Bryozoa, Annelida (Polychaeta), Mollusca in Cnidaria (Anthozoa, Hydrozoa). Avtorji opozarjajo na velik upad morskega travnika kolenčaste cimodoceje v obdobju od 2014 naprej, ki narekuje potrebo po varovanju in upravljanju morskih travnikov v severnem Jadranskem morju.*

Ključne besede: aufwuchs, *Cymodocea nodosa,* morski travnik, sesilni nevretenčarji

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OCCURRENCE OF *GNATHIA* LARVAE (CRUSTACEA, ISOPODA, GNATHIIDAE) IN THREE LESSEPSIAN FISH SPECIES IN THE SOUTHERN TURKISH COAST OF THE AEGEAN SEA

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ABSTRACT

Gnathia *larvae (praniza) of Gnathiidae (Crustacea, Isopoda) were reported for the first time from the southern Turkish coast of the Aegean Sea, found in the gill filaments of Lessepsian species goldband goatfish* Upeneus moluccensis *(Bleeker, 1855) and Red Sea goatfish* Parupeneus forsskali *(Fourmanoir & Guézé, 1976) (both Perciformes; Mullidae), and in the gill filaments and mouth of redcoat* Sargocentron rubrum *(Forsskål, 1775) (Beryciformes; Holocentridae). The prevalence of* Gnathia *larvae in these fish was 47 %, 63 %, and 58 %, mean intensity 1.3, 1 and 1, respectively. The parasites were observed macroscopically in the gill filaments of the fish, and appeared red as the blood sucked from their hosts completely filled their stomachs. A description of morphological characters of the praniza is also provided.*

Key words: *Gnathia* larvae, Goldband goatfish, Redcoat, Red Sea Goatfish, Lessepsian, Turkey

PRESENZA DI LARVE DI *GNATHIA* (CRUSTACEA, ISOPODA, GNATHIIDAE) IN TRE PESCI LESSEPSIANI LUNGO LA COSTA MERIDIONALE TURCA DEL MAR EGEO

SINTESI

Le larve (praniza) di Gnathia *(Crustacea, Isopoda, Gnathiidae) sono state segnalate per la prima volta lungo la costa turca meridionale dell'Egeo, rinvenute nei filamenti branchiali di specie lessepsiane, come la triglia dorata* Upeneus moluccensis *(Bleeker, 1855)* e Parupeneus forsskali *(Fourmanoir & Guézé, 1976) (entrambi Perciformes; Mullidae), e nei filamenti branchiali e nella bocca del pesce armato rosso* Sargocentron rubrum *(Forsskål, 1775) (Beryciformes; Holocentridae). La prevalenza delle larve di* Gnathia *in questi pesci era rispettivamente del 47 %, 63 % e 58 %, con un'intensità media pari a 1,3, 1 e 1. I parassiti sono stati osservati macroscopicamente nei filamenti branchiali dei pesci e sono diventati rossi quando il sangue succhiato ai loro ospiti ha riempito completamente il loro stomaco. Gli autori forniscono anche una descrizione dei caratteri morfologici delle larve.*

Parole chiave: larve di *Gnathia*, triglia dorata, pesce armato rosso, lessepsiani, Turchia

INTRODUCTION

The goldband goatfish, the Red Sea goatfish and the redcoat are known as native to the Indo-Pacific Ocean and Red Sea. After the opening of the Suez Canal, they have been reported as non-indigenous species in the Mediterranean Sea. The goldband goatfish and the redcoat were reported for the first time by Kosswig (1950), and the red sea goatfish by Çınar *et al*. (2006) in the Turkish coast of the Mediterranean Sea . There are several records about these fish species in Turkey: Gücü *et al*. (1994), Taşkavak *et al*. (1998), Kaya *et al*. (1999), Başusta & Erdem (2000), Torcu & Mater (2000), Bilecenoğlu *et al*. (2002), Öğretmen *et al*. (2005), Sangun *et al*. (2007), Gökçe *et al*. (2010), Ergüden & Turan (2013) for the goldband goatfish; Taşkavak *et al*. (1998), Başusta & Erdem (2000), Torcu & Mater (2000), Taşkavak & Bilecenoğlu (2001), Can *et al*. (2002), Öğretmen *et al*. (2005), Kabaklı & Ergüden (2018) for the redcoat; and Yağlıoğlu & Ayaş (2016), Gürlek *et al*. (2016) for the Red Sea goatfish.

The Gnathiidae display sexual dimorphism. Adult forms are free-living organisms found in oscular cavities of sponges, on various substrates, such as galleries in soft sea floor, in coral crevices, or microcliffs of estuaries (Smit & Basson, 2002, Giannetto *et al*., 2003). Larval forms have three stages, with each stage including two forms: praniza and zuphea. Pranizas are known as temporary haematophagous ectoparasites on fish including elasmobranchs and teleosts (Ferreira, 2011). Zupheas are non-feeding benthic dwellers (Hadfield *et al*., 2008; Ferreira, 2011). Pranizas have been reported from the body surface, gill and mouth cavities, and fins of their hosts. A praniza feeds on the blood and tissue fluids of fish; when its gut is filled with the blood of the host, it goes down to the benthos for meal digestion, and later moults into female or male (Tanaka, 2007; Ferreira, 2011).

Several studies have been carried out about pathological and detrimental effects of praniza larvae on their hosts (Paperna & Zwerner, 1976; González *et al*., 2004; Marino *et al*., 2004; Jones & Grutter, 2005). There are also reports of fish deaths caused by praniza infestations from around the world (Paperna & Por, 1977; Paperna & Overstreet, 1981; Mugridge & Stallybrass, 1983; Patarnello *et al*., 1995).

Lessepsian parasites were reported from the Mediterranean (Merella *et al*., 2016; Özak *et al*., 2012; El-Rashidy & Boxshall, 2012), after the occurrence of 18 Lessepsian parasites were recognized by Zenetos *et al*. (2008). In addition to them, the parasitological surveys show that also native parasites were reported from Lessepsian fish (İnnal *et al*., 2007; Shakman *et al*., 2009; Öktener *et al*., 2010; Boussellaa *et al*., 2016; Merella *et al*., 2016; Bakopoulos *et al*., 2017).

These isopods have previously been reported from host species belonging to different fish families native to Turkey (Akmırza, 2000; Akmırza, 2001; Genç *et al*., 2003; Kırkım *et al*., 2008; Alaş *et al*., 2009). Although the mentioned three Lessepsian fish species have colonized the Mediterranean coasts, the parasites associated with them have not been investigated in detail in Turkey. The present study reports the occurrence of new hosts of praniza of *Gnathia* sp in Turkey, complete with morphological characters.

MATERIAL AND METHODS

Redcoat, *Sargocentron rubrum* (Forsskål, 1775) (Beryciformes; Holocentridae) (n = 63), goldband goatfish, *Upeneus moluccensis* (Bleeker, 1855) (Perciformes; Mullidae) (n = 42) from Fethiye Bay (16°17′ N 120°12′ E) and Red Sea goatfish, *Parupeneus forsskali* (Fourmanoir & Guézé, 1976) (Perciformes; Mullidae) (n = 48) from Datça Bay (16°17′ N 120° 12′ E) were caught by gill nets in the Aegean Sea, Turkey in July 2019. The collected parasite samples were fixed in 70% ethanol. Some of the praniza were put in lactic acid for clearing for a minimum of 24 h. The praniza were dissected out in lactic acid between slide and cover slip using Wild M5 and Leica M140 stereo microscopes. All drawings were made with the aid of a drawing tube (Olympus BH-DA) attached to the compound microscope. Measurements are given in millimeters. Identifications and comparisons were performed according to Smit & Basson (2002), Giannetto *et al*. (2003), Hadfield *et al*. (2008) and Ferreira (2011). Scientific names and synonyms of parasites were checked in WoRMS Editorial Board (2020), and fish hosts described according to Froese & Pauly (2019).

RESULTS

Order Isopoda Latreille, 1817
Suborder Cymothoida Wägele, 1989
Superfamily Cymothooidea Leach, 1814
Family Gnathiidae Leach, 1814
Genus *Gnathia* Leach, 1814 (Figs. 1-8, Tab. 1)

Tab. 1: Infestation information concerning praniza.
Tab. 1: Podatki o okužbi s pranico.

Hosts	**Prevalence (%)**	**Mean Intensity**	**Infestation site**
Parupeneus forsskali	63	1	the gill filaments
Upeneus moluccensis	47	1.3	the gill filaments
Sargocentron rubrum	58	1	the gill filaments, mouth cavity



Fig. 1: Praniza of gnathiid isopod. Scale bar: 0.5 mm.
Sl. 1: Ličinka pranica raka enakonožca iz rodu** Gnathia**. Merilo: 0,5 mm.

The trunk colour of the larvae was reddish in appearance as their bodies were filled with the blood of the host. In addition, excess mucus was observed in the gill filaments of the hosts. Infestation information of the praniza is provided in Table 1.

Description of praniza larva (Figs. 1-8): Total body length of praniza larvae is 1.93-2.52 mm, body width 0.61-0.67 mm (n=25). Cephalosome sub-circular and conical-shaped. Posterior margin straight and slightly wider than anterior margin. Cephalon slightly wider than long. Lateral margins slightly convex and parallel. Eyes oval-shaped, large, well developed on lateral margins of cephalosome. Length of eyes is two-thirds of the length of cephalosome. Many melanospots randomly covering dorsal surface of cephalosome. Antenna longer than antennule. Antennule (Fig. 2a) with three peduncle articles, third article longest. Flagellum with four articles, article 2 longest. Both articles 2 and 3 with one and two simple setae; article 4 with one aesthetic seta and five setae. Antenna (Fig. 2b) with four peduncle articles; fourth article longest. Lateral margins of articles 2-4 denticulated. Flagellum with seven articles; article 1 slightly longer. Article 7 with four long setae on distal tip; each article with 1-5 setae on distal end. Mandible (Fig. 2d) stout with swollen basis; distal tip styliform with 12 backwardly directed teeth on its inner margin. Maxillule (Fig. 2e) long and styliform; with eight small teeth on distal inner margin. Maxilla not visible. Maxilliped (Fig. 2c) large, cylindrical, composed of basis and three articled palps. Basis with a long seta and style-like endite. Article 1 with 10 teeth, article 2 with four setae, article 3 with three setae. Gnathopods (Fig. 2f) smaller than pereopods, with seven articles; basis and coxa without pectinate or seta; basis, ischium, merus, carpus, propodus with pectinate scales on inner margins; ischium, merus, carpus, propodus with one seta; dactylus hook-like with a small tooth on medium. Merus the largest article, carpus the smallest article. Paragnath (Fig. 2g) three-segmented, basal segment bears one seta.

Pereopods (Fig. 3) 1-2 similar in size; pereopods gradually increasing from 1 to 5, pereopod 1 being

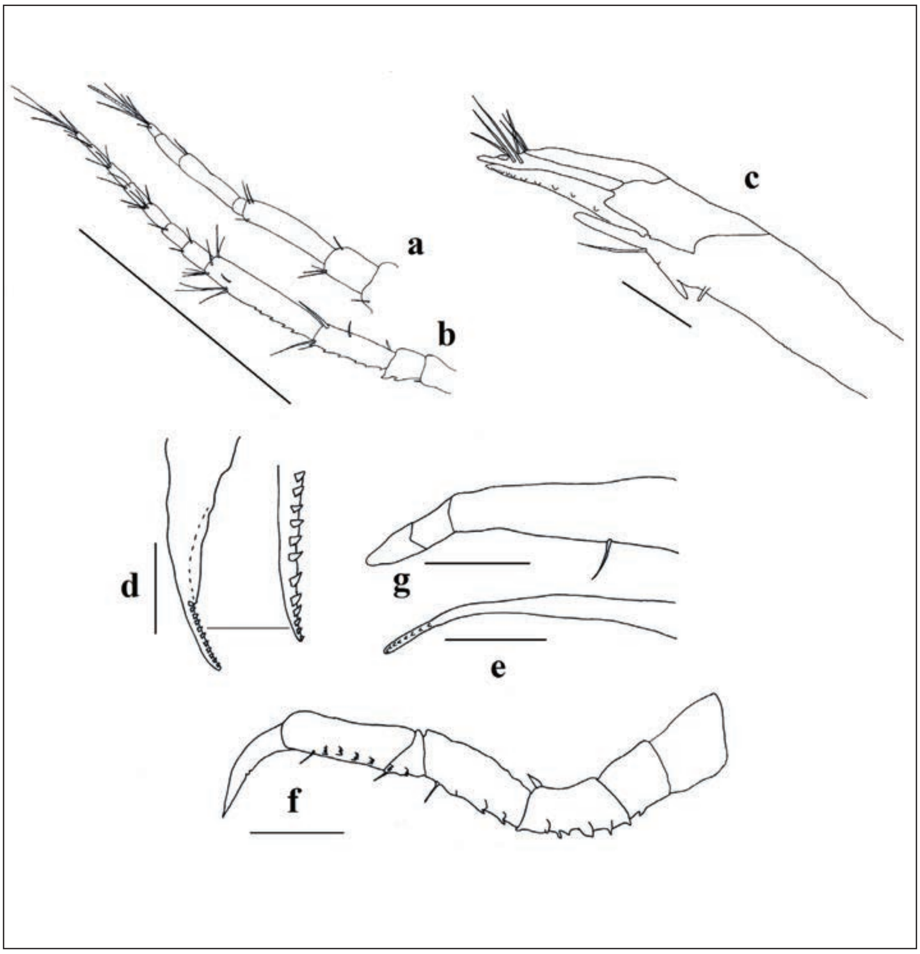


Fig. 2: a. Antennule, b. Antenna, c. Maxilliped, d. Mandible, e. Maxillule, f. Gnathopod, g. Paragnath. Scale bar a & b: 0.38 mm, c & g: 0.05 mm, d: 0.07 mm, e: 0.08 mm and f: 0.09 mm.
Sl. 2: a. antenula, b. antena, c. maksiliped, d. mandibula, e. maksilula, f. gnatopod, g. paragnat. Merilo a & b: 0,38 mm, c & g: 0,05 mm, d: 0,07 mm, e: 0,08 mm in f: 0,09 mm.

the shortest, pereopod 5 the longest. The length of the basis of pereopod 2 about 4 times the width, basis with three simple setae anteriorly, a single simple seta posteriorly, ischium 0.6 times as long as basis, three setae anteriorly, two setae posteriorly, merus 0.6 times as long as ischium, with anterior bulbous protrusion, two simple setae and a single feather-like seta on bulbous protrusion, two setae on posterior margin, carpus 1.2 times as long as merus, four simple setae on posterior margin, propodus 1.2 times as long as carpus, two simple setae on anterodistal margin, two robust setae and a single seta on posterior margin, dactylus 0.6 times as long as propodus, two setae on posterior side and two setae on median side. Posterior margin on propodus of pereopod 1 denticulated; pereopod 2 non-denticulated; posterior margin of propodus, carpus, merus, ischium of pereopod 3 denticulated; posterior margin of propodus, carpus, merus of pereopods 4-5 denticulated.

Pleopods (Fig. 4a) biramous and fan-shaped; endopod larger than exopod in each pleopod. Endopod of pleopod 1 with two articles and bearing 11 plumose setae, exopod with 10 plumose setae. Pleopods 2-3 with 8 plumose setae on endopod and 9 plumose setae on exopod. Pleopod 5 with 7 plumose setae on endopod and 8 plumose setae on exopod. Peduncles of pleopods with two coupling hooks on inner margin, a single seta on inner margin. Endopod (Fig. 4b) larger than exopod. Endopod slightly extending beyond tip of pleotelson. Endopod and exopod with 9 plumose setae. Outer and inner margins of exopod and endopod with short hair-like setae. Uropodal basis with two simple setae.

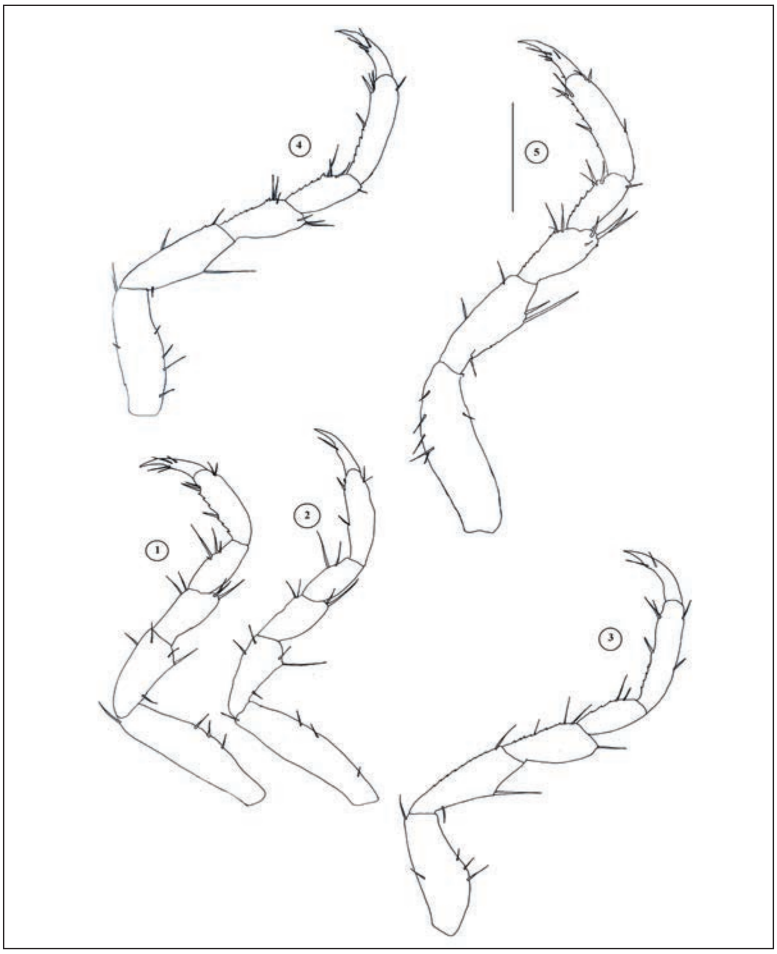


Fig. 3: Pereopods 1-5 of praniza. Scale bar: 0.32 mm.
Sl. 3: Pereiopodi od 1 do 5 pri ličinki pranici. Merilo: 0,32 mm.

DISCUSSION

The fish diversity of Turkish coasts was reviewed by Bilecenoğlu *et al*. (2014), who identified 512 species. Marine fish fauna of Turkey has changed considerably with the arrival of alien species of Indo-Pacific and Atlantic origin through the Suez Canal, Gibraltar, due to climate change, and through ballast water (Oral, 2010; Turan *et al*., 2018). Turan *et al*. (2018) counted 101 non-indigenous fish species reported in Turkish marine waters, including 73 species of Indo-Pacific origin, 22 species of Atlantic origin and 6 species of unknown origin.

The examination of the three Lessepsian fish carried out in this study mainly concerned their length-weight relationships, ecology, and population dynamics in Turkey to date. Although these Lessepsian fishes have been colonizing the Mediterranean coasts since the opening of the Suez Canal in 1869, the parasites associated with them have only been scarcely investigated in the Mediterranean.

Taxonomy of the gnathiid is generally based on the morphology of the free-living male, thus the identification of the morphology of gnathiid larvae is not possible (Smit & Basson, 2002; Hadfield *et al*., 2008; Ferreira, 2011). Hence, the gnathiid larvae in this study could not be identified at species level. There is no previous study providing the description of gnathiid larvae in Turkey. This study is the first report on parasites found in three Lessepsian fish from the Mediterranean Sea.

To date, members belonging to the Gnathiidae have been reported in twenty-eight fish species in the Sea of Marmara, Black Sea, Aegean Sea and Mediterranean coasts of Turkey (Tab. 2). Table 2 provides a general idea about the hosts of Gnathia larvae. Host fish parasitized by Gnathiidae are interpreted according to the feeding type, habitat preference, and family.

There are only a few reports concerning praniza found in members belonging to the Actinopterygii in Turkey. Nunomura and Honma (2004), and Ota

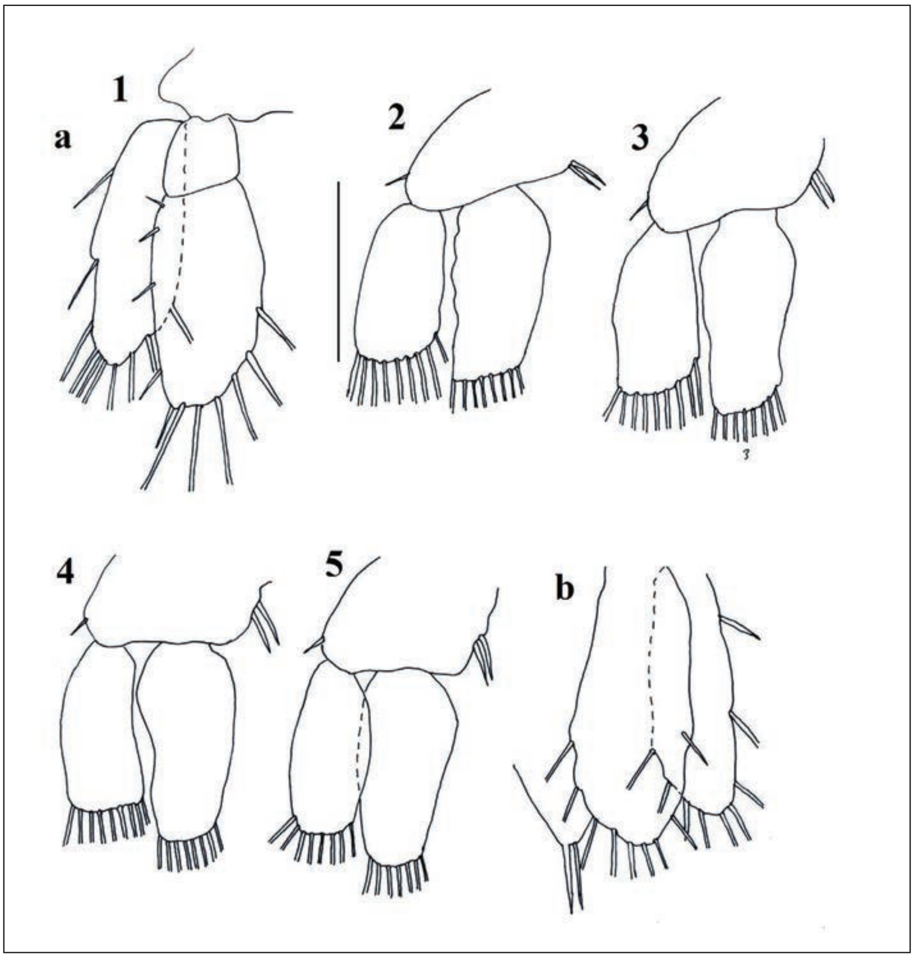


Fig. 4: a. Pleopods 1-5 (0.13 mm), b. uropod. Scale bar: 0.13 mm.
Sl. 4: a. Pleopodi 1-5 (0,13 mm), b. uropod. Merilo: 0,13 mm.

(2015) reported of praniza in elasmobranchs. On the other hand, Mhaisen *et al*. (2018) counted 18 marine fish species (10 bony fishes + 8 cartilaginous fishes) as hosts of *Gnathia* sp. in Iraq. There is no record of praniza in Elasmobranchii in Turkey. Gnathiids are mainly reported in fish belonging to the Sparidae and the Serranidae of the Perciformes in light of the studies carried out in Turkey. Based on the habitat types of the host species, the praniza also seem to display a preference for demersal fish, including reef-associated and benthopelagic, over pelagic fish. When the feeding habits of the host species infested with praniza are examined, it may be said that praniza larvae prefer carnivorous to omnivorous or herbivorous fishes.

The praniza larvae in this study were reported from Lessepsian fishes *U. moluccensis*, *P. forsskali* and *S. rubrum*. The fact that these fish are of demersal character in view of their habitat, and carnivorous by feeding habit, presents them as possible hosts of Gnathiidae praniza larvae.

Bilge *et al*. (2019) analysed the potential invasiveness of 45 Lessepsian marine fishes in the southwestern coasts of Anatolia (Muğla region, Turkey) using the Aquatic Species Invasiveness Screening Kit (AS-ISK). They categorised *Upeneus moluccensis* and *Sargocentron rubrum* as high-risk species, and *Parupeneus forsskali* as a medium-risk species according to both thresholds. After these Lessepsian fishes were first seen in the Mediterranean Sea in 1950, their high-risk potential invasiveness values show that they adapted very well to the Marmara Sea. The fact that these fishes are well settled in the Mediterranean contributes to them being potential hosts of gnathiid praniza.

Although the Gnathiidae are treated as native parasites in this study, it is not known whether this parasite species is invasive. It has been reported from these hosts in the Indian Ocean as well. Chelladurai & Subbulakshmi (2017) reported of gnathiid praniza on *Parupeneus indicus* with a 93.7% prevalence, and *Sargocentron rubrum* with a 63.6% prevalence



Fig. 5: Red Sea goatfish, *Parupeneus forsskali* ***(Fourmanoir & Guézé, 1976), redcoat,*** *Sargocentron rubrum* ***(Forsskål, 1775), goldband goatfish,*** *Upeneus moluccensis* ***(Bleeker, 1855) (top to bottom).***
Sl. 5: *Parupeneus forsskali* ***(Fourmanoir & Guézé, 1976),*** *Sargocentron rubrum* ***(Forsskål, 1775),*** *Upeneus moluccensis* ***(Bleeker, 1855) (od zgoraj navzdol).***



Fig. 6: Praniza on mouth base of redcoat.
Sl. 6: Pranica na ustih veveričevke.

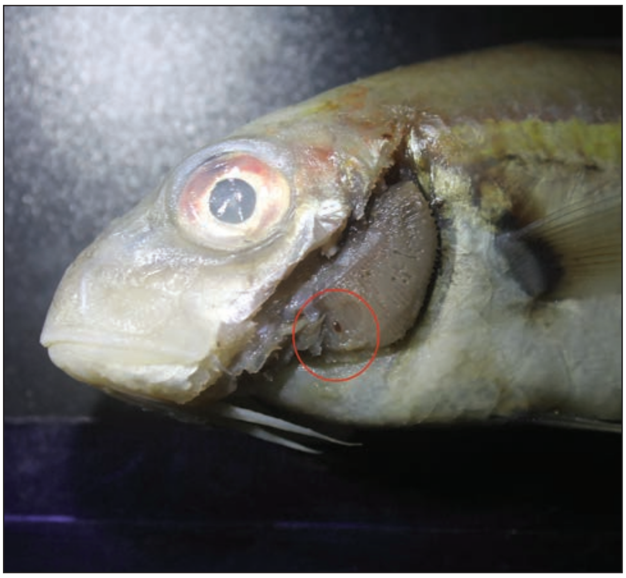


Fig. 7: Praniza on gill filaments of goldband goatfish.
Sl. 7: Pranica na škržnih filamentih vrste *Upeneus moluccensis.*



Fig. 8: Praniza on gill filaments of goldband goatfish.
Sl. 8: Pranica na škržnih filamentih vrste *Upeneus moluccensis**.***

Tab. 2: Reports of gnathiid isopods on fish from Turkey.
Tab. 2: Poročanja o rakih enakonožcih na ribah iz Turčije.

Gnathiid Species	Host	Locality	Record
Praniza larvae	*Diplodus annularis*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Diplodus vulgaris*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Diplodus sargus*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Dentex dentex*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Lithognathus mormyrus*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Pagrus pagrus*	Aegean Sea	Akmırza (2000)
Praniza larvae	*Diplodus annularis*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Diplodus vulgaris*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Symphodus tinca*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Scorpaena porcus*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Scorpaena scrofa*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Gaidropsarus mediterraneus*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Umbrina cirrosa*	Aegean Sea	Akmırza (2001)
Praniza larvae	*Epinephelus aeneus*	Mediterranean Sea	Genç *et al.* (2003)
Praniza larvae	*Epinephelus marginatus*	Mediterranean Sea	Genç (2007)
Praniza larvae	*Ephinephelus costae*	Mediterranean Sea	Erol (2007)
Praniza larvae	*Mullus surmuletus*	Black Sea	Alaş *et al.* (2009)
Praniza larvae	*Scorpaena scrofa*	the Sea of Marmara	Alaş *et al.* (2009)
Praniza larvae	*Serranus cabrilla*	the Sea of Marmara, Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Mugil cephalus*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Gaidropsarus mediterraneus*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Trachurus mediterraneus*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Sarpa salpa*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Diplodus vulgaris*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Sciaena umbra*	Aegean Sea	Alaş *et al.* (2009)
Praniza larvae	*Pagellus erythrinus*	the Sea of Marmara	Alaş *et al.* (2009)
Praniza larvae	*Diplodus annularis*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Diplodus vulgaris*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Lithognathus mormyrus*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Spicara maena*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Pagellus erythrinus*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Coris julis*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Scorpaena scrofa*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Stephanolepis diaspros*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Sparus aurata*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Dicentrarchus labrax*	Aegean Sea	Akmırza (2010)
Praniza larvae	*Conger conger*	Aegean Sea	Akmırza (2012)
Praniza larvae	*Dentex macrophthalmus*	Aegean Sea	Düşen *et al.* (2016)
Paragnathia formica (Hesse,1864)	*Mugil cephalus*	Aegean Sea	Kırkım *et al.* (2008)
Paragnathia formica (Hesse,1864)	*Pagellus erythrinus*	Aegean Sea	Kırkım *et al.* (2008)
Paragnathia formica (Hesse,1864)	*Mugil cephalus*	Mediterranean Sea	Taşkın (2013)

from the southeastern coast of India, Gulf of Mannar. Paperna & Por (1977) reported *Gnathia piscivora* in mullets in Israel. Tuan *et al*. (2015) reported Gnathia sp with a 26.92% prevalence on *Parupeneus multifasciatus* and a 57.14% prevalence on *P. heptacanthus* in Vietnam. Rückert *et al*. (2009) reported of *Gnathia* sp with a 10% prevalence on *Upeneus moluccensis* in Lampung Bay, Indonesia.

Fish parasites have been used to discriminate fish stock in population studies (Avdeev, 1992; MacKenzie & Abaunza, 1998; MacKenzie, 2002; Catalano *et al*., 2014; Poulin & Kamiya, 2015) and in other fields of study, e.g., as pollution indicators (MacKenzie *et al*., 1995; MacKenzie, 1999; Palm & Dobberstein, 1999; Williams & MacKenzie, 2003) since the 1950s.

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POJAVLJANJE LIČINK VRSTE IZ RODU *GNATHIA* (CRUSTACEA, ISOPODA, GNATHIIDAE) PRI TREH LESEPSKIH SELIVKAH V JUŽNIH TURŠKIH VODAH EGEJSKEGA MORJA

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POVZETEK

Avtorji poročajo o prvem pojavljanju ličink (pranica) vrst iz rodu Gnathia *(Crustacea, Isopoda) iz južnih turških voda Egejskega morja, najdenih na filamentih lesepskih bradačev* Upeneus moluccensis *(Bleeker, 1855) in* Parupeneus forsskali *(Fourmanoir & Guézé, 1976) (oba Perciformes; Mullidae) ter na škržnih filamentih veveričevke* Sargocentron rubrum *(Forsskål, 1775) (Beryciformes; Holocentridae). Pojavljanje ličink iz rodu* Gnathia *pri teh ribah je bilo 47 %, 63 %, in 58 %, povprečna intenzivnost pa 1.3, 1 in 1. Zajedavci so bili opaženi na škržnih filamentih in so bili rdeče barve, saj so s krvjo gostitelja povsem zapolnili želodec. Avtorja podajata tudi popis morfoloških znakov pranice.*

Ključne besede: ličinke iz rodu *Gnathia*, *Upeneus moluccensis*, *Parupeneus forsskali*, *Sargocentron rubrum*, lesepske selivke, Turčija

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ADDITIONAL RECORD OF THE ALIEN CRAB *ACTAEODES TOMENTOSUS* (BRACHYURA: XANTHIDAE: ACTAEINAE) FROM TUNISIAN MARINE WATERS

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ABSTRACT

The occurrence of the Xanthid crab Actaeodes tomentosus*, an Erythrean species, is recorded for the second time from the Tunisian marine waters. A single female was captured by hand on October 2016 during scuba diving survey carried out in the Marine Protected Area of Zembra. The specimen was caught at 1 m depth anchored to a rock. The carapace length and width were respectively 9.1 mm and 14.3 mm. This record constitutes the northernmost extension range of the species not only in Tunisia but also at a Mediterranean scale.* A. tomentosus *is neurotoxic containing the "Tetrodotoxin", which is widespread in the Indo-Pacific region and considered among the most common intertidal coral reefs species.*

Key words*:* Marine Protected Area, bioinvasion, decapods, diving survey, extension range, Tunisia

NUOVA SEGNALAZIONE DEL GRANCHIO ALIENO *ACTAEODES TOMENTOSUS* (BRACHYURA: XANTHIDAE: ACTAEINAE) IN ACQUE MARINE DELLA TUNISIA

SINTESI

La presenza di una specie eritrea di granchi della famiglia Xanthidae, Actaeodes tomentosus*, è stata registrata per la seconda volta nel mare della Tunisia. Una singola femmina è stata catturata con le mani nell'ottobre del 2016, durante un'indagine subacquea condotta nell'area marina protetta di Zembra. Il granchio è stato catturato a 1 m di profondità, mentre si trovava saldamente aggrappato ad una roccia. La lunghezza e la larghezza del carapace erano rispettivamente di 9,1 mm e 14,3 mm. Questo ritrovamento costituisce la segnalazione più settentrionale delle specie, non solo in Tunisia, ma anche su scala mediterranea.* A. tomentosus *è una specie neurotossica, contenente la tetrodotossina. È diffusa nella regione indo-pacifica ed è considerata tra le più comuni specie di barriere coralline intertidali.*

Parole chiave: area marina protetta, bioinvasione, decapodi, indagine subacquea, estensione, Tunisia

INTRODUCTION

Invasive species are considered as a major threat (Azzurro *et al.*, 2019) and the second most common cause of species extinction after habitat destruction (Bellard *et al.*, 2016). The Mediterranean is the most invaded Sea in the world (Galil *et al.*, 2014), and of the 821 Non-Indigenous Species (NIS) recorded up to date, more than a half are established (Zenetos *et al.*, 2017). This region is known to support several crab invasions (Swart *et al.*, 2018). Over 39 alien Brachyura species of Red Sea/Indo-Pacific origin have been well documented in the Mediterranean Sea, mainly in the eastern Basin (Klaoudatos & Kapiris, 2014). Decapods are the best known marine invasive crustaceans due to their easy larvae dispersal (Landeira *et al.*, 2019), their high reproductive rate and wide environmental tolerance allowing an important establishment success (Gothland *et al.*, 2014) and particularly the adverse environmental and socio-economic impacts that can inflict such species. Indeed, some crabs from Portunidae (*Portunus segnis* (Forskål, 1775), *Callinectes sapidus* Rathbun, 1896) or Epialtidae (*Libinia dubia* H. Milne Edwards, 1834) families have strongly affected human health, ecosystems and fishery activities in Tunisian waters (Khamassi *et al.*, 2019; Rjiba *et al.*, 2019; Chaffai *et al.*, 2020).

According to Corsini-Foka & Kondylatos (2015), four alien xanthid crabs occur in the Mediterranean Sea; *Atergatis roseus* (Rüppell, 1830), *Actaea savignii*

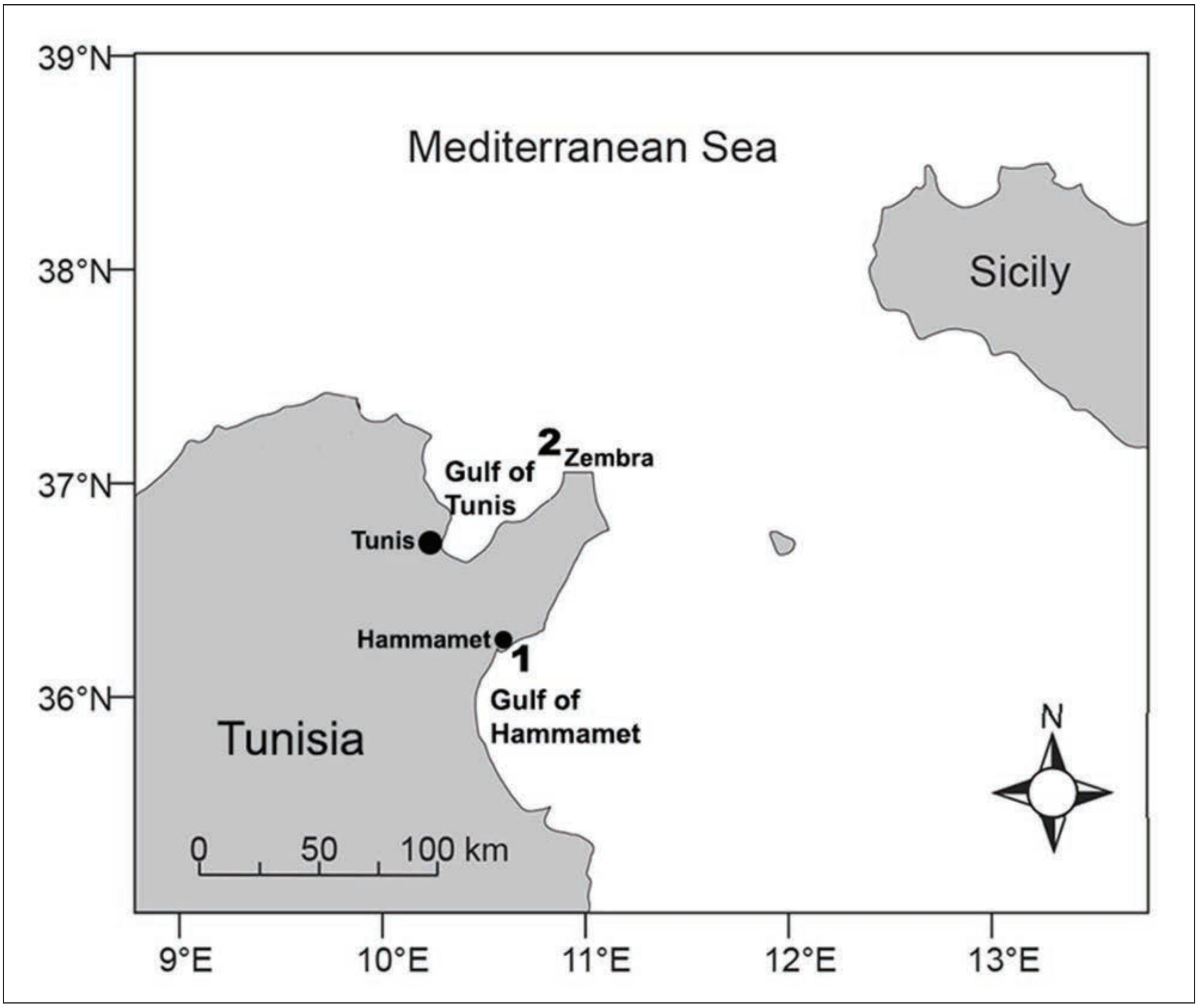


Fig. 1: Map of northern Tunisia (redrawn from Capapé** et al., **2020) indicating the capture sites of** Actaeodes tomentosus **(ref. INAT- IXAN-Ac-tom0) from the Tunisian waters. 1. Marina of Hammamet. 2. Zembra Island.
Sl. 1: Zemljevid severne Tunizije (prerisano iz Capapé in sod., 2020) z lokaliteto, kjer je bila najdena rakovica vrste** Actaeodes tomentosus **(ref. INAT- IXAN-Ac-tom0) v tunizijskih vodah. 1. Marina v Hammametu. 2. Otok Zembra.

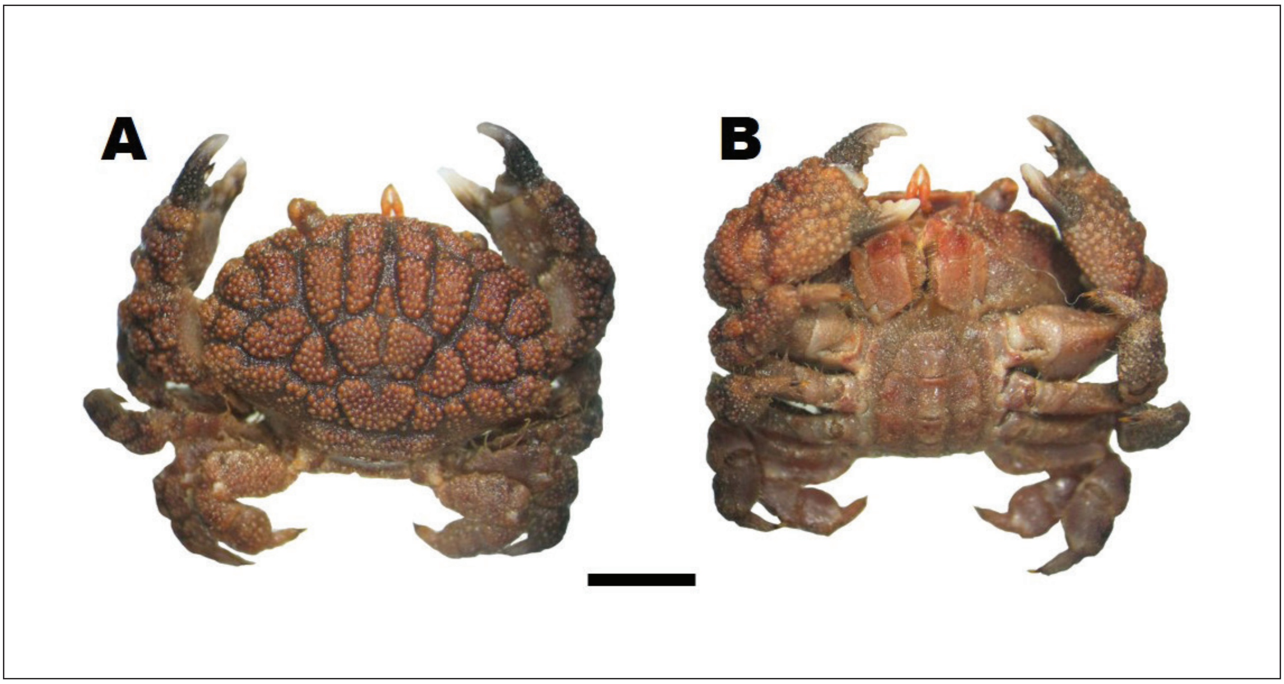


Fig. 2: *Actaeodes tomentosus* ***(ref. INAT- XAN-Ac-tom01). A. Dorsal view. B. Ventral view. Scale bar = 5 mm.***
Sl. 2: *Actaeodes tomentosus* ***(ref. INAT- XAN-Ac-tom01). A. Hrbtna stran. B. Trebušna stran. Merilo = 5 mm.***

(H. Milne Edwards, 1834), *Xanthias lamarckii* (H. Milne Edwards, 1834) and *Actaeodes tomentosus* (H. Milne Edwards, 1834), this latter is the most frequent species, worldwide distributed particularly in the Indo-Pacific region, from the Red Sea, Aden, Somalia, Kenya, Tanzania, Mozambique and S. Africa to the Western Indian Ocean islands up to Australia, Japan and Hawaii Islands (Serène 1984).

A. tomentosus was recorded for the first time in the Mediterranean Sea in the shallow coastal waters of Rhodes Island (Corsini-Foka & Kondylatos, 2015). In January 2015, three specimens, two non-ovigerous females and one male, were collected from the marina of Hammamet, located in eastern Tunisia among biofouling (Ounifi Ben Amor *et al.,* 2016).

A larger trend in recorded alien species in the last decades have been locally reported as a main consequence of a heavy biological invasion of mixed origins. More than 150 NIS fauna have been recorded at present. Among them crustacean decapods constitute the main group of alien fauna, more than 50% according to Ounifi-Ben Amor *et al.* (2016) and Ben Souissi *et al.* (2019).

In order to implement effective management plans for marine ecosystems, an accurate and updated spatio-temporal data on species biogeography is required (Katsanevakis *et al.*, 2020). Records of "New-Comers" should not be limited to their first observation. In fact, species distribution knowledge constitute a prerequisite to assess their invasion potential and its progress and therefore the establishment of the best conservation measures.

MATERIAL AND METHODS

During a periodic assessment of climate change impacts on marine biodiversity carried out mainly in Tunisian Marine Protected Areas (MPA's), a single female of *Actaeodes tomentosus* specimen was collected by hand at 1 m depth anchored to a rock during scuba diving survey on 15 October 2016 performed in Zembra MPA (37°04'645''N and 11°02'960''E) (Fig.1). The crab was identified as *A. tomentosus* following Serène (1984) and was subsequently preserved in 95% alcohol, and deposited in the Collection of crustacean species at the Institut National Agronomique de Tunisie under the catalogue number: INAT- XAN--Ac-tom01.

RESULTS AND DISCUSSION

The collected specimen was a female exhibiting the typical morphological characters of *A. tomentosus* following Serène (1984). The crab was easily identified by numerous dark granules throughout the carapace and appendages, especially on the outer face of both movable and fixed dactyls. (Fig. 2). The carapace length (CL) and width (CW) were respectively 9.1 mm and 14.3 mm. The ratio CW/CL= 1.58 is near the value (1.55) reported for the species by Serène (1984).

A. tomentosus has been reported in the marina of Hammamet on 2015 (Ounifi-Ben Amor *et al*., 2016), suggesting that the possible pathway of introduction was maritime traffic. Since this species has now been found in two localities from eastern (Central Mediterranean) and northeastern Tunisia (Western Mediterranean) for a short time, this record constitutes the northernmost extension range of the species not only in the area but also in the wide Mediterranean Sea. Such patterns suggest that viable populations are progressively establishing although it is small and cryptic, and probably escaped to notice.

The arrival of poisoning species is increasingly noted in Tunisian waters (Yahia *et al*., 2013; Ben Souissi *et al*., 2014; Ounifi-Ben Amor & Ghanem In Dailianis *et al*. (2016). Indeed, cases of intoxication by Tetradotoxin (TTX), present in Tetraodontiformes fish (pufferfish) were observed in western Tunisia during year 2013 (Ben Souissi *et al*., 2014) and also identified in some *A. tomentosus* from Taiwan (Ho *et al*., 2006), and Saxitoxin and related compounds (STXs) were found in specimens from Japan (Deeds *et al*., 2008). Several scientific studies confirm the increasing spread and abundance of invasive marine species of human health concern, however, information on their impacts remains unequally and poorly known (Galil, 2018).

Regular monitoring programs in and around MPAs enhance NIS detection to an early invasion stage probably allow their eradication (Otero *et al*., 2013). However, the risks that NIS can have in these particular habitats are very significant and even harmful (Galil, 2019). Several Non-Indigenous species have been reported the waters surrounding Zembra Island MPA, and such habitat constitute a kind of refuge, where NIS do not face to fishing pressure occurring in these unprotected sites (Giakoumi *et al*., 2019). In fact, and according to Ounifi-Ben Amor *et al*. (2016) and Ounifi-Ben Amor & Ghanem In Dailianis *et al*. (2016), of the 137 non-indigenous faunal species recorded in Tunisia, 25 species were listed around Zembra MPA and six species have been observed in its surrounding waters. Therefore, in total agreement with Galil (2019), it appears that MPAs with an abundance of Non-Indigenous populations could be considered as seed banks leading to a spill-over effect to neighboring areas.

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NOVI ZAPIS O POJAVLJANJU TUJERODNE RAKOVICE *ACTAEODES TOMENTOSUS* (BRACHYURA: XANTHIDAE: ACTAEINAE) IZ TUNIZIJSKIH MORSKIH VOD

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POVZETEK

Avtorji poročajo o drugem zapisu o pojavljanju rakovice Actaeodes tomentosus*, eritrejske vrste, v tunizijskih morskih vodah. Samica te vrste je bila ujeta z roko oktobra 2016 med potapljanjem z avtonomno potapljaško opremo v morskem zavarovanem območju Zembra. Primerek je bil ujet na 1 m globine, tesno pritrjen na skalo. Dolžina oklepa je bila 9,1 mm, njegova širina pa 14,3 mm. Ta zapis predstavlja najsevernejši primer pojavljanja vrste v Tuniziji in tudi v Sredozemskem morju.* A. tomentosus *je nevrotoksična vrsta, ki vsebuje strup tetrodotoksin. Je široko razširjena vrsta v Indo-Pacifiku in pogosta vrsta v bibavičnem pasu na koralnih grebenih.*

Ključne besede*:* morsko zavarovano območje, bioinvazija, raki deseteronožci, potapljaški pregled, širjenje areala, Tunizija

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ADDITIONAL RECORD OF GOLANI ROUND HERRING, *ETRUMEUS GOLANII* (OSTEICHTHYES: DUSSUMIERIIDAE) FROM TUNISIAN WATERS WITH COMMENTS ON ITS DISTRIBUTION IN THE MEDITERRANEAN SEA

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ABSTRACT

The occurrence of the Lessepsian migrant Golani round herring Etrumeus golanii *(Dussumieriidae) was confirmed off the Tunisian coast with the record of a specimen captured by a commercial purse-seiner on 13 March 2020, in the Gulf of Hammamet. The specimen, a mature female, measured 262 mm in total length and weighed 178.22 g. Morphometric and meristic characteristics of the specimen caught are given. This new finding of E. golanii confirms the rapid expansion of this Lessepsian migrant's range in the Mediterranean Sea.*

Key words: Round herring, *Etrumeus golanii*, Lessepsian migration, Gulf of Hammamet, expansion range, Mediterranean Sea

NUOVE SEGNALAZIONI DELLA SARDINA DI GOLANI, *ETRUMEUS GOLANII* (OSTEICHTHYES: DUSSUMIERIIDAE) IN ACQUE DELLA TUNISIA CON COMMENTI SULLA DISTRIBUZIONE NEL MEDITERRANEO

SINTESI

La presenza della sardina di Golani, Etrumeus golanii *(Dussumieriidae), migrante lessepsiano, è stata confermata al largo delle coste tunisine con la cattura di un esemplare con rete da circuizione commerciale, il 13 marzo 2020, nel Golfo di Hammamet. Il campione, una femmina matura, misurava 262 mm di lunghezza totale e pesava 178,22 g. Nell'articolo vengono fornite le caratteristiche morfometriche e meristiche dell'esemplare catturato. Questo nuovo ritrovamento di E. golanii conferma la rapida espansione di questo migrante lessepsiano nel mare Mediterraneo.*

Parole chiave: sardina di Golani, *Etrumeus golanii*, migrazione lessepsiana, Golfo di Hammamet, intervallo di espansione, Mediterraneo

INTRODUCTION

The Golani round herring *Etrumeus golanii* DiBattista, Randall & Bowen, 2012 is a Lessepsian migrant (*sensu* Por, 1978), native to the western Indian Ocean and common in the Red Sea (Golani & Fricke, 2005). The species migrated through the Suez Canal into the Mediterranean Sea, where it was first recorded in the eastern Levant Basin, off Haifa, and misidentified as the red-eye round herring *Etrumeus teres* (DeKay, 1848) by Whitehead (1963). Successive records occurred in eastern Mediterranean regions, where viable populations are probably established (Golani, 2000, 2005).

After migrating toward western areas, the species was first recorded in the central Mediterranean in the waters surrounding the Lampedusa Island by Falautano *et al*. (2006). It was redefined the Golani round herring *E. golanii* by DiBattista *et al*. (2012) and furtherly recorded with its new taxon in Tunisia (Boussellaa *et al.*, 2016, Rafrafi-Nouira *et al*., 2017), Libya (Shakman *et al.*, 2017), Algeria (Kassar & Hemida *in* Stamouli *et al.*, 2017), and Morocco (Tamsouri *et al*. (2019). In this paper, an additional record of *E. golanii* from the Tunisian coast is reported and the species' distribution throughout the Mediterranean and its status in this sea are discussed.

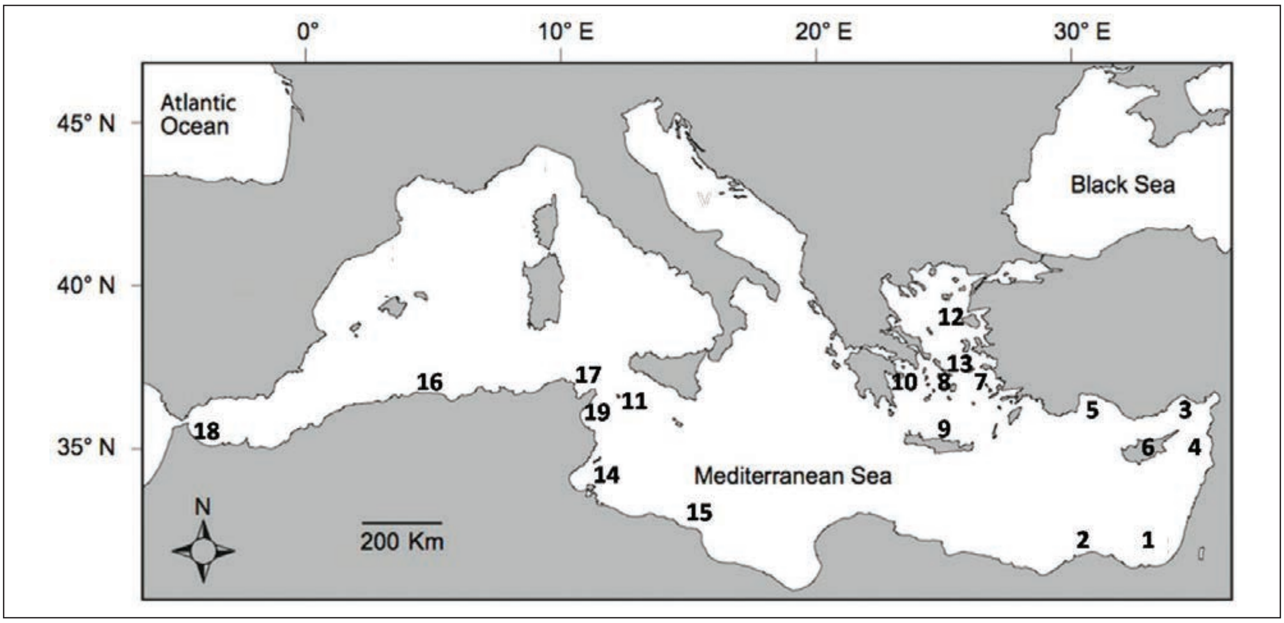


Fig. 1: Map of the Mediterranean Sea, copied from Rafrafi et al. (2015), indicating the distribution of Golani round eye *Etrumeus golanii* ***by chronological order. 1: Haifa Bay, Israel, Whitehead (1963). 2: Off Egypt, El Sayed (1994). 3: Off Iskenderun, Turkey, Başusta et al. (1997). 4: Off Syria, erroneously reported as*** *Etrumeus sadina* ***(Mitchill, 1814), Saad (2002) in Ali (2018). 5: Antalya Gulf, Turkey, Yılmaz & Hoşsucu (2003). 6: Off Limassol, Cyprus, Golani (2005). 7: Rhodes, Greece, Corsini et al. (2005). 8: Cyclades, Greece, Kallianiotis & Lekkas (2005). 9: Hydra Island, Greece, Zenetos et al. (2008). 10: Crete, Greece, Kasapidis et al. (2007). 11: Lampedusa, Italy, Falautano et al. (2006). 12: Dikili Coast, Turkey, Yarmaz et al. (2010). 13: Gulf of Izmir, Turkey, Akyol & Ulaş (2016). 14: Gulf of Gabès, Tunisia, Boussellaa et al. (2016). 15: Off Misrata, Libya, Shakman et al. (2017). 16: Off Cherchell, Algeria, Kassar & Hemida in Stamouli et al. (2017). 17: Off Ras Jebel, Tunisia, Rafrafi et al. (2017). 18: Fnideq Bay, Morocco, Tamsouri et al. (2019). 19: Gulf of Hammamet, Tunisia, present study.***

Sl. 1: Zemljevid Sredozemskega morja (prirejeno po Rafrafi in sod., 2015) in podatki o razširjenosti vrste *Etrumeus golanii* ***glede na časovni interval pojavljanja. 1: Zaliv Haifa, Izrael, Whitehead (1963). 2: v vodah Egipta, El Sayed (1994). 3: v vodah okoli Iskenderuna, Turčija, Başusta in sod. (1997). 4: pred Sirijo, napačno določena kot*** *Etrumeus sadina* ***(Mitchill, 1814), Saad (2002; v Ali 2018). 5: zaliv Antalya, Turčija, Yılmaz & Hoşsucu (2003). 6: pred Limassolom, Ciper, Golani (2005). 7: Rodos, Grčija, Corsini in sod. (2005). 8: Kikladi, Grčija, Kallianiotis & Lekkas (2005). 9: otok Hydra, Grčija, Zenetos in sod. (2008). 10: Kreta, Grčija, Kasapidis in sod. (2007). 11: Lampedusa, Italija, Falautano in sod. (2006). 12: obala Dikili, Turčija, Yarmaz in sod. (2010). 13: zaliv Izmir, Turkey, Akyol & Ulaş (2016). 14: zaliv Gabès, Tunizija, Boussellaa in sod. (2016). 15: pred Misrato, Libija, Shakman in sod. (2017). 16: v vodah pred Cherchell, Alžirija, Kassar & Hemida v Stamouli in sod. (2017). 17: pred Ras Jebel, Tunizija, Rafrafi in sod. (2017). 18: zaliv Fnideq, Maroko, Tamsouri in sod. (2019). 19: zaliv Hammamet, Tunizija, pričujoča raziskava.***

MATERIAL AND METHODS

On 13 March 2020, a specimen of *E. golanii* was observed at the fish landing site of Kélibia. The specimen was caught in the Gulf of Hammamet at 36°46′57″ N and 11°17′52″ E, during the night, at a depth of 40-42 m (Fig. 1). It was caught together with other pelagic teleost species such as the round sardinella *Sardinella aurita* Valenciennes 1847, the Mediterranean horse-mackerel *Trachurus mediterraneus* (Steindachner, 1868), and the Atlantic chub mackerel *Scomber colias* Gmelin, 1789. The specimen of *E. golanii* was frozen and delivered to the laboratory, where it was identified, photographed, measured and weighed. Morphometric and meristic characteristics were recorded following Nielsen & Johnson (1983), and are summarized in Table I. The specimen was preserved in 10% buffered formaldehyde, deposited in the Ichthyological Collection of the Institut Supérieur de Pêche et d'Aquaculture de Bizerte (Tunisia), under catalogue number ISPAB-Etr-gol-01.

RESULTS AND DISCUSSION

The specimen of *E. golanii* from eastern Tunisian waters measured 262 mm in total length (TL) and weighed 178.22 g (Fig. 2). The specimen was identified as *E. golanii* based on a combination of morphological characteristics: body elongated and cylindrical in its anterior part, large head, eye covered by adipose eyelid, dorsal fin origin before midpoint, pelvic fin behind dorsal fin base, a single W-shaped pelvic scute at the base of pelvic fins, lack of series of scutes along the belly, scales very deciduous, easily detached, colour dark blue with silvery flanks and belly.

The present specimen was as an adult female at stage 4 of maturity, with 4.13 g of gonad weight. Such observation is in agreement with Yarmaz *et al.* (2010), Boussellaa *et al.* (2016), Falautano *et al.* (2006) and Tamsouri *et al.* (2019). Size at first sexual maturity for *E. golanii* from the Egyptian Mediterranean waters ranged from 122 to 126 mm in males and from 120 to 131 mm in females (El-Sayed 1996, Osman *et al.* 2013). Additionally, most of the individuals caught so far have been relatively large adults (Yarmaz *et al.,* 2010).

The stomach of this specimen was empty. Boussellaa *et al.* (2016) noted that *E. golanii* fed on preys similar to those previously reported, such as zooplankton, mainly copepods and euphausiids rather than fish larvae and molluscs (Froese & Pauly, 2005; Kallianiotis & Lekkas, 2005; Osman *et al.*, 2013; Tanaka *et al.*, 2006). These observations suggest that *E. golanii* has probably adapted to its new environment.

Morphometric and meristic characters of *E. golanii* were recorded and all data are in total agreement with those from other areas of the central and eastern Mediterranean Sea (Table 1). Therefore, the present finding constitutes the third record of this species in Tunisian waters, based on a single specimen. Seven specimens were captured in southern Tunisia, Gulf of Gabès, and one specimen, described by Rafrafi-Nouira *et al.* (2017), was singled out from a haul of specimens captured in northern Tunisia, off Ras Jebel, by pelagic trawl following an experienced fisherman aware of local fishing grounds. Additionally, fishermen from north-eastern Tunisia landing at the fishing site of Kélibia reported that this species is occasionally observed together with *S. aurita* and *S. colias*. Therefore, the occurrence of *E. golanii* in the Tunisian coast cannot be ruled out, however further records are needed before confirming the successful local establishment of the species.



Fig. 2: Specimen of *Etrumeus golanii* ***(ref. ISPAB Etr-gol-01) captured in the Gulf of Hammamet, Tunisia, scale bar = 20 mm.***
Sl. 2: Primerek vrste *Etrumeus golanii* ***(ref. ISPAB Etr-gol-01), ujet v zalivu Hammamet, Tunizija, merilo = 20 mm.***

Tab. 1: Morphometric measurements and meristic counts recorded for the specimen of Etrumeus golanii specimen caught in the Gulf of Hammamet (ref. ISPAB Etr-gol-01), and those recorded for specimens captured in other Mediterranean areas.

Tab. 1: Morfometrične meritve in meristična štetja pri primerku vrste Etrumeus golanii, ujetem v zalivu Hammamet (ref. ISPAB Etr-gol-01) in pri primerkih, ujetih v drugih predelih Sredozemskega morja.

Authors	**Cyprus (Lymassol) 2000** **Golani (2000)**	**Italy (Lampedousa) 2006** **Falautano *et al.* (2006)**	**Turkey (Dikili Strait) 2009** **Yarmaz *et al.* (2009)**	**Turkey (Izmir Bay) 2016** **Akyol & Ulaş (2016)**	**Tunisia (Gulf of Gabes) 2014** **Boussellaa *et al.* (2016)**	**Tunisia (Ras Jebel) 2017** **Rafrafi-Nouira *et al.* (2017)**	**Morocco (Alboran Sea) 2018** **Tamsouri *et al.* (2019)**	**Tunisia (Gulf of Hammamet) 2020** **Present study**
Number of specimens	2	1	1	1	7	1	7	1
				Measurements (mm)				
Total length	-	231	149	180	225–265	222	252–283	262
Fork length	-	211	138	159	200–243	200	228–260	236
Standard length	138–213	202	127	153	165–225	188	215–243	224
Body depth	20.6–42.4	35.6	25	28	34.5–41.3	41.2	43–50	48
Predorsal fin length	—	88	59	66	87–102	82.5	92–107	98
Prepectoral fin length	—	—	—	38	—	—	46.5–56	52
Preanal fin length	—	—	—	127	—	—	175–203	186
Head length	31–53.5	45	19	34	39.2–49.3	44.3	45–52	50
Eye diameter	9.3–18.6	12.44	9	11	10.2–12.5	13	13–15	14
Preorbitary length	—	—	—	12	—	—	13–16	15
Dorsal fin base length	—	26.6	17	—	24.2–26.1	31.6	31–35	34
Anal fin base length	—	9.4	9	—	9–9.5	11	9–12	11
Pelvic fin length	—	14.3	—	—	14–22	14	14–16	15
				Meristic counts				
Dorsal fin rays	17–20	18	18	17	18	19	18	18
Pectoral fin rays	15–17	15	16	16	16	16	15–16	17
Pelvic fin rays	8–10	8	7	8	8	9	8	8
Anal fin rays	9–10	12	9	9	9	9	9–10	9

An update of the currently known distribution of this species in the Mediterranean is summarized in Figure 1. It is evident that *E. golanii* is abundantly recorded in the eastern Mediterranean, where viable populations have probably established. Additionally, the species constitutes an important local commercial resource (Akyol & Ulaş, 2016, Corsini *et al.,* 2005, DiBattista *et al.,* 2012). The presence of *E. golanii* received a positive feedback without any negative impact on local fisheries resources, as reported by Kassar & Hemida in Stamouli *et al.* (2017) and Tamsouri *et al.* (2019).

In conclusion, the recent observation of an additional specimen of *E. golanii* in the central Mediterranean Sea (Tunisian waters) may be linked to the environmental parameters, which are becoming more favourable for this species (Tamsouri *et al.*, 2019). As a consequence, we could in the future expect the establishment of a viable population in Tunisian waters confirming the expansion of this species throughout the Mediterranean Sea.

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NOV ZAPIS O POJAVLJANJU VRSTE *ETRUMEUS GOLANII* (OSTEICHTHYES: DUSSUMIERIIDAE) IZ TUNIZIJSKIH VODA S KOMENTARJI O NJENI RAZŠIRJENOSTI V SREDOZEMSKEM MORJU

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POVZETEK

V zalivu Hammamet v tunizijskih vodah so 13. marca 2020 komercialni ribiči v zaporno plavarico ujeli primerek vrste Etrumeus golanii *(Dussumieriidae). Bila je samica, ki je merila 262 mm v dolžino in tehtala 178,22 g. O ulovljenem primerku avtorji podajajo morfometrične meritve in meristične podatke. Novi podatek o pojavljanju vrste E. golanii potrjuje hitro razširjanje te lesepske selivke v Sredozemskem morju.*

Ključne besede: *Etrumeus golanii*, lesepska selitev, zaliv Hammamet, širjenje areala, Sredozemsko morje

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ONESNAŽEVANJE OKOLJA

INQUINAMENTO DELL'AMBIENTE

ENVIRONMENTAL POLLUTION

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RESEARCH AND CHARACTERIZATION OF DETERMINANTS CONTROLLING THE ACCUMULATION OF CERTAIN METALS IN THE LEAVES OF *DYSPHANIA AMBROSIOIDES*

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ABSTRACT

In this research, we are interested in the study of the leaves of the plant Dysphasia ambrosioides *and its extracts: on the one hand, by IR spectroscopic analysis, thermogravimetry (TGA), and determination of metals by atomic absorption spectrophotometer (SAA); and on the other, carrying out phytochemical screening of extracts of leaves of* D. ambrosioides*.*

Key words: heavy metals, *Dysphasia ambrosioides*, pollution, alkaloids, tannins, glycosids, flavonoids

RICERCA E CARATTERIZZAZIONE DEI DETERMINANTI CHE CONTROLLANO L'ACCUMULO DI ALCUNI METALLI NELLE FOGLIE DI *DYSPHANIA AMBROSIOIDES*

SINTESI

In questa ricerca gli autori si sono interessati allo studio delle foglie della pianta Dysphasia ambrosioides *e dei suoi estratti: da un lato, mediante analisi spettroscopica IR, termogravimetria (TGA) e determinazione dei metalli mediante spettrofotometro ad assorbimento atomico (SAA); e dall'altro, effettuando lo screening fitochimico di estratti di foglie di* D. ambrosioides.

Parole chiave: metalli pesanti, *Dysphasia ambrosioides*, inquinamento, alcaloidi, tannini, glicosidi, flavonoidi

INTRODUCTION

Today, a significant percentage of the drugs authorized by government agencies are naturally occurring molecules, or compounds derived therefrom (about 50%). As a consequence, there is a significant potential for discovering new molecules of therapeutic interest in plants. Among these plants we chose to study the *Dysphasia ambrosioides* (L) Mosyakin & Clemants. It is a wild species of tropical America naturalized in the Old World, an upright herb, annual or perennial, with a more or less pubescent branching stem. It is commonly employed as an antimicrobial, antifungal (Paul *et al*.,1993; Boutkhil *et al*., 2009; Boutkhil *et al*., 2011; Cicera *et al*., 2018), anti-rheumatic, analgesic (Okuyama *et al*., 1993), sedative, antipyretic (Gadano *et al*., 2006), also used for the treatment of respiratory, urogenital disorders, and vascular, nervous, and metabolic disorders such as diabetes and high cholesterol (Cruz *et al*., 2007), due to its cytotoxic (Ruth *et al*., 2015), antioxidant, anti-inflammatory and anti-Leishmanial activities (Monzotea *et al*., 2014; Luz *et al*., 2017; Reyes-Becerril *et al*., 2019).

Among pollutants generated by industrial activities, heavy metals (i.e., Cu, Pb, Cr, etc.) pose several concerns. These elements readily bio-accumulate and have a recognised eco-toxicity. Moreover, they are involved in several pathologies (in the central nervous system, liver, kidneys; and can also cause cancers and embryonic malformations) (Abrahams *et al*., 2002).

Today, a lot of research investigates the impact of heavy metals on the rate of germination and plant growth. For example, Mihoub *et al*., (2005) showed that during the germination of pea seeds (*Pisum Sativum* (L.)), the cotyledons in stressed grains gradually accumulate Cd and Cu, and retain high contents of Fe, Mg, and Zn. Some plants have little or no tolerance and die in contact with heavy metals. Others have defence reactions, and slow absorption by secreting acids which will increase the pH and consequently reduce the mobility of trace elements. Others are metal tolerant, and even accumulate them, concentrating them. These plants are said to be "hyper-accumulative" and metallophilic. The trace elements are absorbed by the roots and most often stay there. The translocation in the aerial parts (stems, leaves) varies depending on the metal and indicates an increase in the concentration of metals in the soil. Lead remains in the roots, while Cd passes more easily through the aerial parts. Studies have shown that certain plants, called metallophytes, are capable of developing normally on sites highly contaminated with various metals and some of these plants, qualified as hyper-accumulators (Brooks, 1998), are capable of massively storing metals in their aerial parts. There is also phyto-extraction, based on the use of hyper-accumulative plants, which absorb metals from the soil and accumulate them in aerial organs (McGrath, 1998). This method is effective for a wide variety of heavy metals (Pb, Cd, Ni, Zn…).

Phenolic compounds are a widely used class of secondary metabolites and are located in the vacuoles of plant cells, in the intercellular space as well as on the surface of plants. They form a large group of compounds which include simple phenols, such as phenolic acids, flavonoids (flavones, flavonols and anthocyanins) and polymerized phenols, such as tannins and lignins. They participate in defence reactions against pathogens, and in allelopathy protect cell structures from the unwanted effects of excess photochemical energy and ultraviolet radiation, especially UV-B rays. Phenolic compounds found in flowers and fruits have the property to colour these organs (Winkel-Shirley, 2001).

In this research, a preliminary investigation in order to study the leaves of the *D. ambrosioides* plant by determining its chemical composition was conducted by IR spectroscopy, thermogravimetric analysis (TGA), and determination of metals by atomic absorption spectrophotometer (SAA). A phytochemical screening for other compounds present in the aqueous extract was performed as well.

MATERIAL AND METHODS

Collection of samples

The leaves of *Dysphasia ambrosioides* were collected during spring, 2019, at Ain Orma park (33°53′36″N 5°32′50″W), which is located between the cities of Meknes and Khemissat, in the region of Fes-Meknes (Morocco).

After collection, the leaves of the plant were washed separately, dried at room temperature in a dry and ventilated space, and protected from light to avoid loss of active substances. After drying, the various organs were finely ground and powdered using an electric mill. The powder obtained was stored in closed jars and kept in absence of light.

Analytical techniques

Infrared spectroscopy was used to identify the chemical functions of organic molecules. Briefly, the infrared radiation is an electromagnetic radiation with a wavelength greater than that of the visible light but shorter than that of the microwave light. The infrared domain studied was between 4000 cm^{-1} and 400 cm^{-1}, which corresponds to the vibration energy domain of the bonds. The apparatus used in this analysis was Fourier transform infrared spectroscopy (IR-TR) type JASCO 4100.

Thermogravimetric analysis is a thermal analysis technique which consists of measuring the mass of a sample when it is subjected to temperature variations (or time) in an inert environment (Nitrogen, Argon, or Helium for high-temperature tests), or oxidant (dioxygen). The thermogravimetric analysis device (TGA) employed was the Shimadzu thermal analysis type. The curves recorded for

temperature ranged from 0 °C to 700 °C. The heating rate was equal to 10 °C/min.

For mineralization and dosing, 6 g of sample were put in a porcelain dish and calcined at 600 ºC in a muffle furnace (t=6 hours). The ash obtained was mineralized with 75% HNO_3 in a beaker and then brought to dryness until the mineralization discoloured (t=4 hours). The residue was filtered on Whatman-type filter paper.

The determination of heavy metals was carried out using a flame atomic absorption spectrometer (Shimadzu-type model AA-7000). The device was controlled by WIZARD software. A hollow cathode lamp (Hamamatsu Photonics K.K.) was used as the radiation source and a deuterium lamp for the correction of non-specific absorptions. The carrier gas used for the flame was a mixture of air-acetylene. The standard solutions were prepared by diluting the stock solutions with a concentration of 1000 mg/L. The calibration range was prepared according to the element to be assayed.

Several procedures were used to determine the different chemical groups contained in a plant organ. These are tests based on solubility tests, colouring, and precipitation reactions, as well as exams under ultraviolet light.

The quantitative study of the raw extract by means of spectrophotometric assays aimed at determining the total content of total polyphenols, total flavonoids, and condensed tannins. Three calibration curves were drawn for this objective and carried out for each type of assay. The results in gallic acid, quercetin and catechin equivalent are expressed in mg/g of dry matter.

Fifty grams of the powder was added to 500 mL of absolute ethanol, the mixture stirred for 24 hours at 4 °C, then let stand for a few hours. The mixture was then filtered through glass wool and then through sintered glass (funnel N° 03), the filtrate stored at 4 °C until use.

The determination of the total polyphenols was carried out by the Folin-Ciocalteu method described by Wende *et al.*, (2007) with some modifications. This colorimetric method is based on the reduction of the phosphotungsten-phosphomolybdenum complex of Folin reagent by the phenolic groups of the samples, yielding products of blue colouring in alkaline media. Briefly, 0.1 mL of the extract of was added to 2.5 mL of distilled water and 0.5 mL of Folin reagent. After 5 min, 1.0 mL of sodium carbonate (20%) was added to the reaction mixture and the whole incubated for 1 hour at room temperature. The absorbance was read at 765 nm using a UV spectrophotometer. The results are expressed in milligram equivalent of gallic acid/g of dry extract with reference to the calibration curve of gallic acid.

The determination of flavonoids was carried out according to the method of aluminium trichloride ($AlCl_3$) (Bahorun *et al.*, 1996); 1 mL of each extract (prepared in methanol) with suitable dilutions was added to 1 mL of the $AlCl_3$ solution (2% in methanol). After 10 minutes of incubation and reaction, the absorbance was read at 430 nm using a UV spectrophotometer. The results are expressed in mg equivalent of quercetin/g of dry extract with reference to the standard curve for quercetin.

The dosage of condensed tannins was carried out for the extract according to the method of Richard *et al.*, (1978) and Heimler *et al.*, (2006). At 400 µL of each sample or standard (prepared in methanol and in distilled water for Aq. E.) with suitable dilutions, 3 mL of the vanillin solution (4% in methanol) and 1.5 mL of concentrated HCl were added. After 15 min, the absorption was read at 500 nm. The concentration of tannins is deduced from the calibration range established with catechin and expressed in milligrams of catechin equivalent per gram of dry extract (mg EC/mg ES).

RESULTS AND DISCUSSION

The water content in samples is 4.48 %, which is a low value. Several factors could influence the water and dry matter content of the plant, such as the nature of the fibres, the age of the plant, the condition of the soil, and the shelf life of the plant after harvest. The infrared spectroscopic analyses of the calcined samples at temperatures of 110 °C, 325 °C, 450 °C and 600 °C are illustrated in Figure 1.

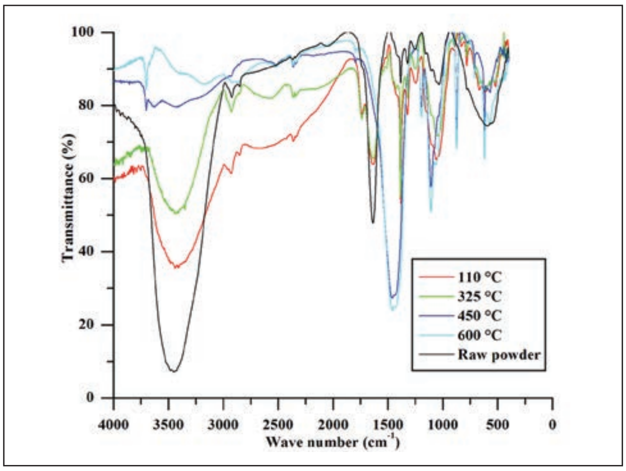


Fig. 1: IR spectrum (KBr) of leaves of** Dysphania ambrosioides **at different temperatures.
Sl. 1: IR spekter (KBr) listov vrste** Dysphania ambrosioides **pri različnih temperaturah.

The IR spectrum shows the presence of a broad and intense band around 3500 cm^{-1} attributable to the valence vibration band of the alcohol function $\nu_{(O-H)}$, and another band which appears around 2900 cm^{-1} attributable to the valence vibration band $\nu_{(C-H)}$. Similarly, we note the presence of a thin band around 1700 cm^{-1} relating to the valence vibration band of $\nu_{(C=O)}$. All of these bands assume that the powder contains organic molecules having alcohol and ketone. Fliou et al., 2019 analysed the

Daphne gnidium L. plant by infrared spectroscopy. The results obtained are similar to those found in this work.

At temperatures of 110 °C and 325 °C, we notice the persistence of the valence vibration bands: $\nu_{(OH)}$, $\nu_{(C\text{-}H)}$, and $\nu_{(C=O)}$ and a decrease in their intensity. At 450 °C, there is the disappearance of two bands relating to the vibration bands of the alcohol and ketone functions, and the appearance of a new band around 1480 cm^{-1} relating to the valence vibration band Ca ($CaCO_3$). This could be explained by the beginning of the disappearance of organic matter. At 600 °C, we notice the disappearance of organic matter, and the appearance of the bands relative to other mineral elements such as kaolinite, smectite, calcite and silicon oxide (Fig. 2) (Hachi *et al.*, 2002).

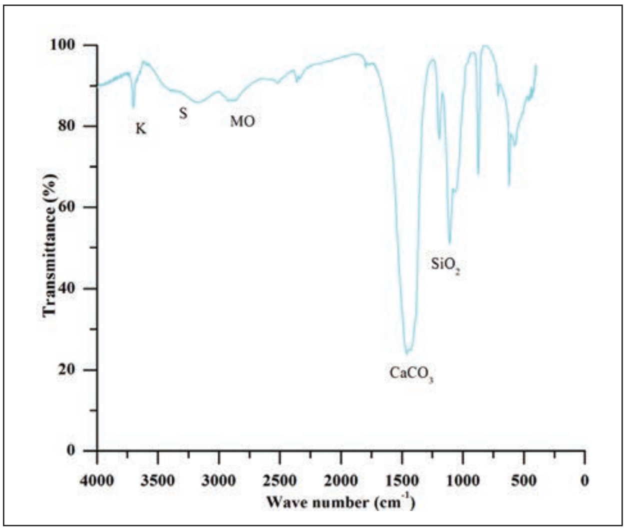


Fig. 2: IR spectrum of powdered leaves of *D. ambrosioides* ***at 600 °C. (K: Kaolinite [$Al_2Si_2O_5(OH)_4$], MO: Organic matter, S: Smectite, Calcite [$CaCO_3$]: Silicon oxide [SiO_2]).***
Sl. 2: IR spekter listov v prahu vrste *D. ambrosioides* ***pri 600 °C. (K: Kaolinit [$Al_2Si_2O_5(OH)_4$], MO: Organska snov, S: Smectit, Kalcit [$CaCO_3$]: Silicijev dioksid [SiO_2]).***

To follow the loss of sample mass during the rise of temperature, we used thermogravimetric analysis (TGA) and differential thermal analysis (DTA). The temperatures related to degradation rates were evaluated. The thermogram obtained is shown in Figure 3.

The thermal degradation of the sample can be identified by the decrease in its weight. The difference in mass is due to the endothermic and exothermic combustion reactions that occur.

The transformation process is characterized by thermal degradation presented by 3 stages: the first corresponds to a mass loss of 0.747 mg or 11.87 %. This loss, which is observed at a temperature of 110 °C, is attributed to

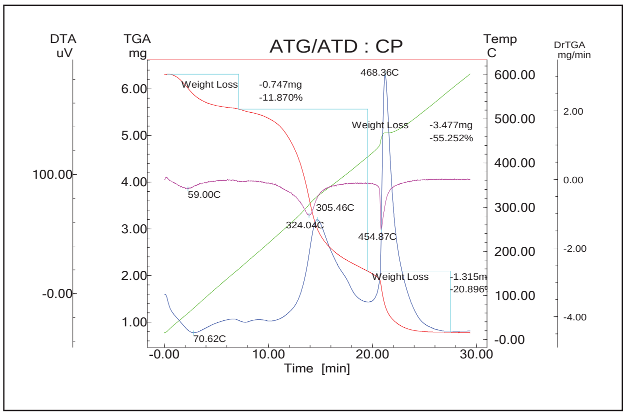


Fig. 3: ATG/ATD curve of the raw powder of the leaves of *D. ambrosioides.*
Sl. 3: ATG/ATD krivulja prahu listov vrste *D. ambrosioides.*

the evaporation of the water contained in the plant. The second step is observed at 325 °C, corresponding to the start of the thermal degradation of organic matter with a mass loss of 3.477 mg or 55.25%. Finally, the third stage, at 454.87 °C, records a mass loss of 1.315 mg or 20.90% and is related to the total destruction of organic matter (Tab. 1).

Tab. 1: Mass loss of leaves of *D. ambrosioides* ***as an effect of temperature.***
Tab. 1: Izguba mase listov vrste *D. ambrosioides* ***pri različnih temperaturah.***

Plant	Step	Temperature	A loss of mass (%)
Powder leaves of *Dysphania ambrosioides*	1	110 °C	11.87
	2	325 °C	67.12
	3	600 °C	88.02

The ATD diagram of the plant shows peaks indicating the different degradation reactions. An endothermic peak at 70.62 °C is attributed to the evaporation of absorbed water and two exothermic peaks, at 324.04 and 468.36 °C, are attributed to the degradation of organic matter. Indeed, the results of the calcinations confirm those of differential thermal analysis (DTA) by the loss of half of the organic matter at a temperature of 300 °C, and that this loss is considerable at 600 °C. The contents of heavy metal are presented in Table 2.

The results revealed a high retention of Na and Ca, with a content of 22.117 and 37.2633 mg/kg, respectively. These concentrations are below the authorized limit. Other elements such as Fe, Cu, Zn and Li are present at low contents, while Cd, Pb, K are almost non-existent. These results show that all the heavy metal contents are

***Tab. 2: Heavy metal content in leaves of** D. ambrosioides.*
***Tab. 2: Vsebnost kovin v listih vrste** D. ambrosioides.*

Metalelement	Content (mg/kg)	Content normal in plants by OMS (mg/kg)	Normal concentration (mg/Kg) (Kabata-Pendias, 1986)	Heavy metal content in the human body (mg/kg) (according to Schroeder, 1967)
Iron (Fe)	1.5175	-	-	60
Copper (Cu)	0.1256	150	-	1
Zinc (Zn)	1.1637	-	27 - 150	33
Cadmium (Cd)	0.003	0.3	0.05 - 0.2	-
Lead (Pb)	0.0145	10	5 - 10	-
Sodium (Na)	22.127	-	-	800
Lithium (Li)	0.1154	-	-	-
Potassium (K)	0.008	-	-	-
Calcium (Ca)	37.2633	-	-	19000

Tab. 3: Results of phytochemical screening of the extract of** D. ambrosioides. **(-): Absence, (+): Presence.
Tab. 3: Tab. 3: Rezultati fitokemičnega pregleda izvlečkov vrste** D. ambrosioides. **(-): Odsotnost, (+): Prisotnost.

			Aqueous extract of the leaves of D. ambrosioides
Alkaloids			+
Tannins	Catechic tannins		+
	Gallic tannins		-
Anthracenederivatives	Free anthracene		-
	Anthracenecombined	O-heteroside	-
		Heterosidegenin	-
		C-heteroside	-
flavonoids	Anthocyanins		-
	Flavones		-
	Flavanones		-
	Flavonols		-
	Leucoanthocyanins		+
	Catechol		-
Saponosides			-
Sterols and tri-terpenes			+++
Mucilage			+
Oses and holosides			+++
Prothocyanidols			-
Iridoids			-

lower than the standards proposed by the WHO, Kabata-Pendias, (1986) and Schroeder, (1967). This suggests that *D. ambrosioides* is not toxic with these trace elements. The results of total polyphenols, total flavonoids, and condensed tannins contents in the aqueous extract are summarized in Table 3.

Photochemical screening revealed the richness in this plant of secondary metabolites, such as alkaloids, catechic tannins, flavonoids (Leucoanthocyans), sterols and tri-terpenes, mucilages, oses and holosides. These results were found by Oliveira *et al.* (2017), who demonstrated that these secondary metabolites found in *D. ambrosioides* have positive effects in the fight against cattle ticks.

The results also show the absence of certain families, such as gallic tannins, anthracene derivatives, anthocyanins, flavones, flavonones, flavonols, catechols, saponosides, prothocyanidols and iridoids. The latter are considered to be powerful allelopathic agents, that is to say that they produce secondary metabolites which can alter the growth and/or the development of other systems (Rodrigues *et al.*, 2009; Lôbo *et al.*, 2008).

The concentration of total polyphenols is based on the regression equation (r^2=0.992) of the calibration range established with gallic acid (Fig. 4). It is expressed in milligrams of gallic acid equivalents per gram of the dry extract (mg EAG/g ES).

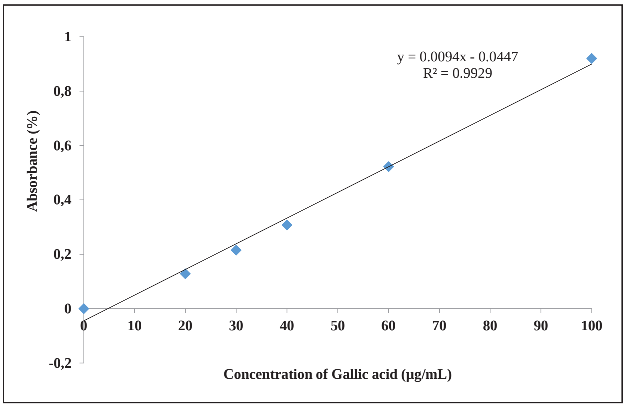


Fig. 4: Gallic acid calibration curve for the determination of total phenols.
Sl. 4: Umeritvena krivulja galne kisline za določevanje celokupnih fenolov

These results suggest that the ethanoic extract is rich in total phenolic compounds, with a content of 42.57 mg EAG/g ES. Previous works (Nowak *et al.*, 2016) on *Chenopodium* (L.) showed that the highest levels of polyphenols were observed in *Chenopodium album* (3.36 mg/g DW), seeds of *Chenopodium urbicum* (3.87 mg / g DW) and *C. urbicum* roots (1.52 mg/g DW). According to Dini *et al.*, (2010) the seeds of bitter *Chenopodium quinoa* contain

86.4 mg of AGE/10 g DW and of sweet *C. quinoa* 77.2 mg of AGE/10 g DW. *Chenopodium pallidicaule* has a higher total polyphenol content of 413 mg GAE/100 g DW (Dasgupta *et al.*, 2007).

The concentration of flavonoids was deduced from the calibration ranges established with quercetin (Fig. 5). It is expressed in milligrams of quercetin equivalent per gram of the dry extract (mg EQ/g ES). According to the calibration curve, the total content of flavonoids extracted from the extract of *D. ambrosioides* leaves with ethanol is of the order of 20.19 (mg EQ/g ES).

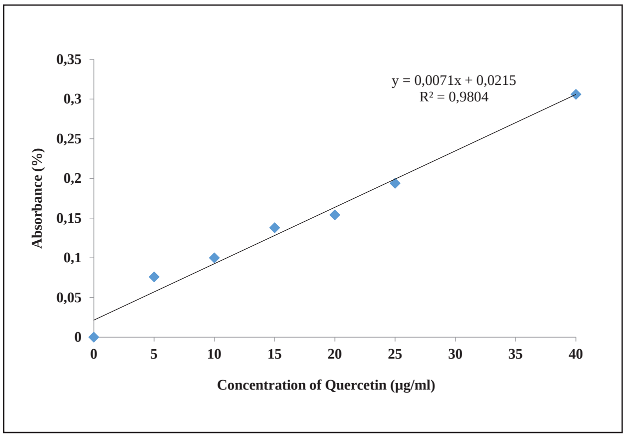


Fig. 5: Quercetin calibration curve for the assay of flavonoids.
Sl. 5: : Umeritvena krivulja za kvercetin za analizo flavonoidov.

The concentration of flavonoids was determined using the spectrophotometric method in the presence of aluminium chloride. The results obtained showed that the concentration of flavonoids in the extract is 20.19 mg EQ/g of ES. This value is lower than the value found by Tanzeel *et al.*, (2018), either a content of 57±1.41 µgQE/mg of extract. Sajjad *et al.*, (2016) have explained this variation in phenolic and flavonoid compounds in different parts of the plant by the polarity of the solvent and with antioxidant and medicinal properties. The calibration curve was constructed using catechin as a reference standard (Fig. 6).

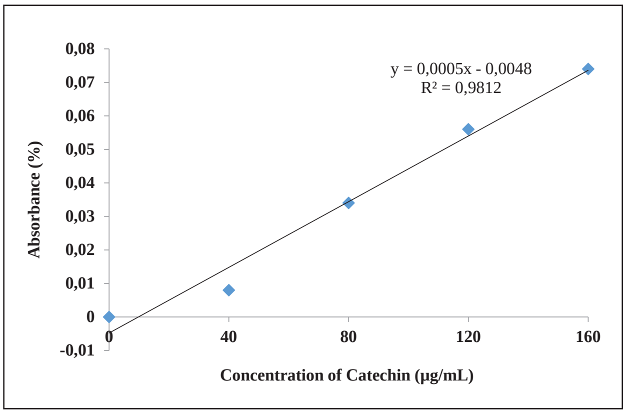


Fig. 6: Catechin calibration curve for the determination of condensed tannins.
Sl. 6: Umeritvena krivulja katehina za določevanje kondenziranih taninov.

According to the calibration curve, the content of condensed tannins in the extract of leaves of *D. ambrosioides* is of the order of 38.78 (mg EQ/g ES). The results showed that the content of condensed tannins in the ethanoic extract of the leaves of *D. ambrosioides* is 38.78 mg EQ/g ES. Upon comparison of these results with those of Ksouri *et al.* (2009) on *Tamarix gallica*, whose leaves recorded a total activity of 14.66 mg EAG/g DM, we note that the content of condensed tannins in the ethanoic extract of the leaves of *D. ambrosioides* can be considered to have strong antioxidant activity because they are very good scavengers for free radicals and also inhibit the formation of superoxide radicals.

CONCLUSIONS

Infrared spectroscopic analysis and differential thermal analysis have shown that the plant *D. ambrosioides* undergoes a loss of organic matter as the temperature increases. The remaining mineral matter was analysed by atomic absorption spectroscopy. The results showed that the plant contained certain metallic elements in small quantities, such as Na, Ca, Fe, Cu, Zn and Li, while the content of Cd and Pb was almost non-existent. Phytochemical screening of the extracts showed a significant presence of total polyphenols, total flavonoids and condensed tannins.

RAZISKAVA O DEJAVNIKIH, KI VPLIVAJO NA KOPIČENJE NEKATERIH KOVIN V LISTIH VRSTE *DYSPHANIA AMBROSIOIDES*

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POVZETEK

V pričujoči raziskavi so avtorji raziskovali liste rastline Dysphasia ambrosioides *in njihove izvlečke z uporabo IR spektroskopske analize, termogravimetrije (TGA), določali kovine z atomsko absorpcijsko spektrofotometrijo (SAA) ter opravili fitokemični pregled izvlečkov listov vrste* D. ambrosioides.

Ključne besede: težke kovine, *Dysphasia ambrosioides*, onesnaženje, alkaloidi, tanini, glikozidi, flavonoidi

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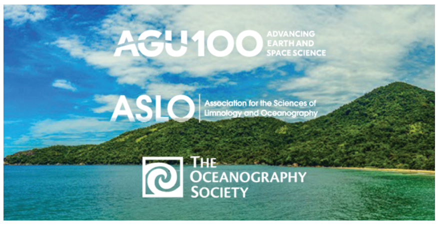
DELO NAŠIH ZAVODOV IN DRUŠTEV

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OCEAN SCIENCES MEETING | 16–21 FEBRUARY 2020 SAN DIEGO, CA, USA



SREČANJE ZNANOSTI O OCEANIH (OCEAN SCIENCE MEETING - OSM)

Ameriški združenji za geofiziko (AGU), za znanost limnologije in oceanografije (ASLO) in Društvo za oceanografijo (TOS) so organizirali največjo letošnjo konferenco raziskovalcev in drugih zainteresiranih, ki se ukvarjajo z vodami, rekami, jezeri, estuariji, podtalnimi vodami, morji in oceani. Srečanje Znanosti o oceanih (Ocean Science Meeting - OSM) je potekalo v centru San Diega, ZDA, od 17. do 20. februarja 2020. Moto letošnjega srečanja je bil »For a Resilient Planet« (Za vzdržljiv planet) in poziv, da lahko le partnerstvo raziskovalcev, z vladami in javnostjo, omogoči ohranjanje zdravih oceanov, zagotavlja varno in trajno preskrbo s hrano ter pomaga ublažiti posledice podnebnih sprememb. Združeni narodi so obdobje od 2021 do 2030 označili za »Desetletje znanosti o oceanu za trajnostni razvoj«, da bi spodbudili mednarodno usklajevanje in sodelovanje v raziskovalnih in znanstvenih programih za boljše upravljanje z viri oceanov in obalnih območij ter zmanjšali nevarnosti.

Srečanja se je udeležilo 6300 raziskovalcev iz 66 držav, od tega je bilo kar 32 % študentov. Žal je srečanje odpovedalo veliko število raziskovalcev zaradi širjenja korona virusa. Raziskovalni dosežki so bili predstavljeni v 16 različnih sekcijah, s predavanji (1820 raziskovalcev) ali posterji (3233 posterjev). Sekcije so vključevale vsebine s področij fizikalne oceanografije, morske ekologije, biodiverzitete, biologije in biogeokemije, mikrobiologije in molekularne ekologije, morske geologije, sedimentologije, kot tudi predstavitve rezultatov prognostičnih modelov, procesov v obalnih morjih in estuarijih, interakcij zrak - morje, fizikalne in biološke interakcije, razvoja opazovalnih sistemov, inštrumentov in senzorjev, zakisevanja, hipoksij in posledic klimatskih sprememb, kot tudi socio-ekonomske vidike in izobraževanje.

Srečanje se je pričelo s plenarnim predavanjem Nainoa Thompson, havajskega domorodca, ki je po starem polinezijskem izročilu izdelal kanu Hōkūleʻa in opremljen s tradicionalnimi navigacijskimi aparaturami jadra po svetu. Kot predsednik Polinezijskega združenja za plovbo, neprofitne raziskovalne in izo-



Fig. 1: Letošnjo Watsonovo nagrado za življenjsko delo je prejela Heidi Sosik (Woods Hole - WHOI).

braževalne organizacije, je pred kratkim zaključil štiriletno potovanje po svetu, kjer se je srečeval z ljudmi, svetovnimi voditelji in predaval kako pomembno je, da ohranjamo kulturno dediščino, skrbimo za naravne vire, ohranjamo zdrav ocean in ščitimo ogrožena območja.

Združenja vsako leto podelijo vrsto nagrad za odmevne raziskovalne dosežke in življenjsko delo. Letošnjo nagrado za življenjsko delo (Watsonovo nagrado) je prejela Heidi Sosik (Woods Hole - WHOI), ki se ukvarja z ekologijo mikroorganizmov, njihovo morfološko in taksonomsko raznolikostjo in interakcijami med organizmi. S sodelavci je razvila podvodni avtomatski sistem za opazovanje in snemanje mikroskopskih organizmov, ki predstavljajo pomemben člen v prehranski verigi, sodelujejo pri izmenjavi plinov in s tem vplivajo na zemeljsko klimo, občasno pa povzročajo škodljiva cvetenja in s tem vplivajo na zdravje ljudi. Erik van Sebille, oceanograf z Univerze Utrecht za projekt Tracking of plastic in our Sea (TOPIOS), ki proučuje poti prenosa/širjenja plastike v oceanih s ciljem, da ustvari 3D zemljevid onesnaženja oceanov s plastiko in omogoči sledenje do izvora. Nagrado Ocean Science Meeting 2020 za prepoznaven prispevek mladega raziskovalca v preteklem letu je prejela Cristina Romera-Castillo (Institut de Ciències del Mar, CSIC) za odmeven članek o visokih koncentracijah raztopljenega organskega ogljika v oceanu, ki so posledica sproščanja organskih spojin s plastike. Sybil Seitzinger (Univerza v Viktoriji) je bila nagrajena za dolgoletne dosežke na področju limnologije in oceanografije na področju raziskav, izobraževanja in ozaveščanja. Medaljo Mary Sears je prejela Jane Lubchenco (Oregon State University) za objave inovativnih in pomembnih raziskav na področju biološke oceanografije in za izjemne prispevke k izobraževanju in mentorstvu na tem področju. Nagrada AGU Sverdrup Award Lecture je prejela Mary-Louise Timmermans (Univerza Yale) za odlične prispevke o atmosferi in oceanih ter za spodbujanje sodelovanja pri atmosferskih in oceanografskih raziskavah.

Zaključno predavanje je podala Margaret Leinen, direktorica Scripps inštituta za oceanografijo. Leinen je prejemnica številnih nagrad, z bogatimi nacionalnimi in mednarodnimi izkušnjami na področju oceanske znanosti, globalnih podnebnih in okoljskih vprašanj, sicer pa paleo-oceanografijna in paleo-klimatologinja. Na raziskovalnem področju je proučevala predvsem sedimente oceanov in biogeokemične



Fig. 2: Farooq Azam in Ake Hägstrom med obiskom Morske biološke postaje Piran Nacionalnega inštituta za biologijo.

cikle, spremembe oceanov in podnebja. Predsedovala je Nacionalni znanstveni fundaciji (NSF) in neposredno vplivala na nekatere najpomembnejše programe na področju morskih znanosti ter povezav z atmosfero in kopnim. V svojem predavanju je predstavila časovni niz objav in poročil ključnih raziskav, strokovnih srečanj in pozivov raziskovalcev, ki so privedli do sprejetja dokumenta Združenih narodov Desetletje znanosti oceanov za trajnostni razvoj (2021-2030). Tako je postavljen okvir prioritetnih področij, nujna je mednarodna koordinacija in sodelovanja, ki omogočajo okrepitev raziskovalnih zmogljivosti na področju morskih ved in prenosa tehnologij.

Na srečanju smo raziskovalci z Morske biološke postaje Piran Nacionalnega inštituta za biologijo sodelovali s prispevki v različnih sekcijah. Alenka Malej s prispevkom o spremembah zooplanktona v toplejšem in bolj oligotrofnem morju, Valentina Turk s predstavitvijo rezultatov patogenih bakterij z naslovom »The adaptation of selected pathogenic microbes to elevated temperature and their detection *In Situ* in the coastal marine environment«, ter mlada raziskovalka Neža Orel s predstavitvijo rezultatov z naslovom »Response of the ambient marine microbial community to a mixture of pollutants in the coastal ecosystem«.

V prostornem kongresnem centru, ki sprejme 125.000 obiskovalcev in ga je načrtoval kanadski arhitekt Arthur Erickson, je 57200 m^2 dvoran in razstavnega prostora. Na OSM je sodelovalo več kot 350 razstavljavcev 122 različnih podjetij in 55 lokalnih in mednarodnih novinarjev. Potekale so številne predstavitve različnih mednarodnih projektov.

Po dvajsetih letih sem ponovno obiskala San Diego, slikovito obmorsko mesto v Kaliforniji, ki mu daje poseben pečat bližina Mehike in ime Scripps, ki se pojavlja na številnih oglasih, imenih podjetjih, raziskovalnih in izobraževalnih inštitutov. Ellen Browning Scripps je bila ameriška žurnalistka, solastnica največje časopisne verige v Ameriki in filantropistka, ki je s svojimi donacijami podpirala nekatere velike inštitute v Južni Kaliforniji. Po konferenci smo se udeležili srečanja bivših študentov in sodelavcev ob obletnici izjemnega raziskovalnega opusa prof. Farooq Azama na Scripps inštitutu za oceanografijo. Kot Fulbright štipendistka sem imela priložnost, da sem leta 2000 sodelovala z njim in njegovimi raziskovalci. Srečanja so se udeležili vidni raziskovalci na področju morske mikrobne ekologije in virologije, med njimi Jed Furman, Ake Hägstrom, Meinhard Simon, Forest Rohwer, Lihini Aluwihare, Koji Hamasaki, Kay Bidle, Mayali Xavier in Brian Palenik.

Farooq Azam je raziskovalec in profesor morske mikrobiologije na oddelku za Morsko biologijo in Centru za morsko biotehnologijo in biomedicino, Scripps inštituta za oceanografijo, Univerze v Kaliforniji, San Diego. Azamove primarne raziskave so s področja ekologije morskih bakterij in virusov, njihove raznolikosti in populacijske dinamike. S sodelavci je bil tvorec teorije mikrobne zanke, v začetku 80 let. Njegove študije vključujejo biokemične in molekularne prilagoditve bakterij na življenje v oceanskem okolju, pomen bakterij in virusov v ciklu oceanskega ogljika, v procesih razgradnje organske snovi in v strukturi in delovanju prehranskih spletov v oceanu. Je prejemnik številnih nagrad in odlikovanja kot je UCSD Excellence in Research Award (1997), nagrada Plymouth Marine Sciences Partnership (1996), G. Evelyn Hutchinson Medalja Ameriškega društva za limnologijo in oceanografijo (1995), medalja Rosenstiel za oceanografske znanosti Miami's School of Marine and Atmospheric University iz Miamija (1984). Leta 2004 je bil Azam izvoljen v Ameriško akademijo za mikrobiologijo in isto leto prejel »honoris causa«, častni doktorat na Univerzi Kalmar na Švedskem in od mednarodnega društva za mikrobiološko ekologijo prejel inauguracijsko Tiedje nagrado za izjemne dosežke na področju mikrobne ekologije. Farooq Azam je večkrat obiskal Morsko biološko postajo Piran, z njim smo skupaj z italijanskimi in hrvaškimi kolegi sodelovali na projektu CREICO ter nato tudi v okviru bilateralnega projekta. Počastil nas je tudi z uvodnim predavanem ob otvoritvi mednarodnega simpozija mikrobne ekologije SAME11 (Symposium on Aquatic Microbial Ecology), leta 2009, ki se ga je udeležilo 210 domačih in tujih raziskovalcev iz 30 različnih držav. Simpozij je organizirala Morska biološka postaja Piran Nacionalnega inštituta za biologijo, ob svoji 40-letnici, potekal pa je pod pokroviteljstvom takratnega predsednika države dr. Danila Türka.

Valentina Turk
Morska biološka postaja Piran, Nacionalni inštitut za biologijo

OCENE IN POROČILA

RECENSIONI E RELAZIONI

REVIEWS AND REPORTS

OCENE IN PRIPOROČILA, 129–129

Book review: A MINIATURE OCEAN
Authors: Lovrenc Lipej, Manja Rogelja & Borut Mavrič
Editor: High school, electrical and naval school Piran (GEPŠ), 221 pp.

I was asked to prepare my scientific opinion regarding the manuscript of the monograph entitled "*A miniature ocean*". This manuscript is dealing with the Aquarium in Piran, an institution with long tradition in the Slovenian coastal town. The monograph with 222 pages is a remarkable compilation of data, concerning the Piran Aquarium and its scientific and educational role in the northern Adriatic area. Opening chapters offer a short description of the history of aquaristics and the roles of modern aquariums. The authors are trying to convey the importance of aquaria from different aspects, such as marine biology, nature conservation, education, cultural role and popularization. Separate chapter is dealing with the scientific contribution the staff of the Piran Aquarium made together with the partner institutions, such as the Marine Biology Station Piran of the National Institute of Biology, in publishing in scientific literature, with special regards to invasive species, tropicalization, and rare, less known and endangered species. A special chapter is dedicated to the functioning of the Piran Aquarium from various aspects, such as animal collection and husbandry, life support systems, etc. The bulk of the monography presents a survey of algal and animal species, which are regularly or occasionally displayed in the tanks of Piran Aquarium. One hundred and seventy species are presented in this chapter. A key for understanding various definitions is presented before the list itself, so the reader has no problem in understanding what certain labels mean. Every single species is presented on one page with a close-up photograph, a short description, its size, habitat and distribution. Finally, if there are any facts the reader may find interesting, they are mentioned at the very end of the page. This chapter is followed by a dictionary where all scientific terms are explained, by index of Latin and Slovenian names of species, literature and presentation of the authors. The monograph is illustrated with more than 230 excellent color photographs and some original illustrations in black and white.

Since books such as this one are not common in scientific literature, it is quite difficult to compare it and asses its value. However, it is a valuable contribution since it presents in detail an important Slovenian institution and shows the importance such institution has to the local community, also demonstrating the wide range of activities done by the staff of the Aquarium. One of the most important tasks is certainly raising the awareness of how rich the Adriatic Sea actually is.



To my opinion the value of this book is in detailing various contributions the Aquarium has made to the scientific community and providing an overview of marine turtles rehabilitation cases. The monography is also trying to present the importance of new processes such as bioinvasion and tropicalization in modifying the floral and faunal communities in the northern Adriatic Sea.

The survey of aquarium species is also a valuable part, since it could provide help to professionals working in the field of education.

The monography "*A miniature ocean*" is to my opinion a valuable contribution as it presents finer points of good aquarium practice to a wider public, while also turning the reader's attention to the richness and diversity of the Adriatic Sea.

Milena Mičić
Director of Aquarium Pula

KAZALO K SLIKAM NA OVITKU

SLIKA NA NASLOVNICI: Zajedavci so pomemben, a velikokrat spregledan vidik morske biodiverzitete. Pogosto se zgodi, da na ribjih gostiteljih najdemo vrste zajedavcev, ki so slabo poznane ali zelo redke. Takšen je tudi primer zajedavskega ceponožca vrste *Demoleus heptatus*, ki je bil najden na primerku morskega psa šesteroškrgarja *(Hexanchus griseus)* v Izoli januarja 2018. (Foto: D. Trkov)

Sl. 1: Pranica na posnetku je ličinka rakov enakonožcev iz družine Gnathiidae. Ličinka je zajedavka na raznih vrstah obrežnih rib. (Foto: D. Trkov)

Sl. 2: Zajedavski raki ceponožci iz rodu *Caligus* zajedajo številne vrste rib. Na svetu jih živi več kot 220 različnih vrst, med njimi tudi takšne, ki povzročajo gospodarsko škodo. (Foto: D. Trkov)

Sl. 3: Endoparazite najdemo znotraj ribjega gostitelja. Čeprav nekateri povzročajo škodo na komercialno pomembnih ribah, je o njihovi biologiji le malo znanega. To velja tudi za sesače (Trematoda). (Foto: D. Trkov)

Sl. 4: Trije predstavniki rakov ceponožcev iz rodu *Caligus*, od katerih imata dva jajčne filamente, so bili najdeni na velikem prisesniku (*Lepadogaster candoli*i). (Foto: D. Trkov)

Sl. 5: Ribja uš, predstavnica skupine zajedavskih rakov enakonožcev iz družine Cymothoidae, je vidna za očesom dolgonosega morskega konjička (*Hippocampus guttulatus*). (Foto: L. Lipej)

Sl. 6: Morski travniki kolenčaste cimodoceje (*Cymodocea nodosa*) so pomembna življenjska okolja, ki nudijo veliko ekosistemskih servisov. V zadnjem desetletju raziskovalci poročajo o tem, da se morski travniki soočajo z drastičnim krčenjem. (Foto: L. Lipej)

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FRONT COVER: Parasites are an important, although often neglected part of marine biodiversity. Findings of rare and less-known parasites in fish hosts are frequent. Such was also the case of the parasitic copepod *Demoleus heptatus* found in the bluntnose sixgill shark (*Hexanchus griseus*) in Izola (Slovenia) in January 2018. (Photo: D. Trkov)

Fig. 1: Praniza is the larval stage of marine isopods of the family Gnathiidae. It parasitizes various species of coastal fish. (Photo: D. Trkov)

Fig. 2: Parasitic copepods of the genus *Caligus* infest many fish species. There are more than 220 different species known, some of them causing substantial economic damage. (Photo: D. Trkov)

Fig. 3: Endoparasites live inside the bodies of their fish host. Although some represent a threat to commercially important fish, their basic biology remains poorly investigated. That is also true of trematodes. (Photo: D. Trkov)

Fig. 4: Three copepods of the genus *Caligus*, two of them with filamentous egg strings, were found in the Connemara clingfish (*Lepadogaster candolii*). (Photo: D. Trkov)

Fig. 5: The cymothoid sea louse, a representative of the group of parasitic isopods, can be seen behind the eye of the long-snouted sea horse (*Hippocampus guttulatus*). (Photo: L. Lipej)

Fig. 6: Seagrass meadows of *Cymodocea nodosa* are important habitats, known to provide many different ecosystem services. Over the past decade, scientists have reported that seagrass meadows are faced with a drastic shrinkage of their coverage. (Photo: L. Lipej)

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